



Parasitoid Diversity Associated with Dipteran Larvae and Pupae in Animal Feces and Organic Substrates: A Review of Experimental Studies

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Abstract: Parasitoids are important natural enemies of dipterans, acting on different developmental stages, particularly larvae and pupae, in environments associated with decomposing organic matter and animal feces. This study aimed to review experimental works on parasitoid species associated with dipteran immatures collected from different substrates, including bovine and chicken feces, carcasses, and organic waste, with emphasis on parasitoid diversity, host associations, substrate influence, and ecological factors affecting parasitism. The results revealed a high diversity of parasitoid species, including *Brachymeria podagrica* (Fabricius, 1787) (Hymenoptera: Chalcididae), *Hemencyrtus herbertii*, Ashmead 1900 (Hymenoptera: Encyrtidae), *Nasonia vitripennis* (Walker, 1836) (Hymenoptera: Pteromalidae), *Pachycrepoideus vindemmiae* (Rondani, 1875) (Hymenoptera: Pteromalidae), *Paraganaspis egeria* Díaz, Gallardo & Walsh, 1996 (Hymenoptera: Figitidae), *Spalangia* spp. (Hymenoptera: Pteromalidae) and *Trichopria* sp. (Hymenoptera: Diapriidae), which parasitize different developmental stages of dipterans, reflecting distinct biological strategies and ecological niches. Substrate type strongly influenced parasitoid occurrence, with higher diversity and activity observed in environments rich in organic matter, such as carcasses and animal feces. Environmental factors, including temperature, humidity, and stage of substrate decomposition, also played an important role in determining host availability and parasitoid activity. The integration of data from multiple studies indicates that parasitoid occurrence is closely associated with host presence and environmental conditions. These findings highlight the ecological importance of parasitoids in regulating dipteran populations and support their potential application in biological control programs targeting synanthropic flies. Further research is needed to improve the understanding of parasitoid–host interactions across different substrates and environmental conditions.

Keywords: Biological Balance, Insecta, Natural Enemies, Substrate, Traps.

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RESEARCH PAPER

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How to cite this paper:

Carlos Henrique Marchiori &
Klebert de Paula Malheiros (2026).
Parasitoid Diversity Associated
with Dipteran Larvae and Pupae in
Animal Feces and Organic
Substrates: A Review of
Experimental Studies. *Middle East
Res J Biological Sci*, 6(3): 159-199.

Article History:

| Submit: 22.04.2026 |

| Accepted: 25.05.2026 |

| Published: 27.05.2026 |

1.0. INTRODUCTION

Dipteran species are widely distributed in different environments and are commonly associated with decomposing organic matter, where they participate in nutrient cycling and ecological balance. However, several species also have sanitary, veterinary, and economic importance, because they can transmit pathogens and cause myiasis in humans and animals. *Chrysomya albiceps* (Wiedemann, 1819) (Diptera: Calliphoridae) and *Musca domestica* L., 1758 (Diptera: Muscidae) are examples of synanthropic species frequently associated with animal carcasses, feces, organic waste, and other decomposing substrates (Marchiori, 2019a; Marchiori, 2021a; Guimarães *et al.*, 2026; Österman *et al.*, 2026).

Some dipteran species may also influence the structure of communities associated with decomposing

substrates by competing with or preying on other immature insects. In this context, parasitoids are important natural enemies because they develop in or on their hosts and usually cause host death. These insects are commonly associated with larvae or pupae of flies in different substrates, and their diversity and occurrence may vary according to host availability, substrate type, environmental conditions, and seasonality (Marchiori, 2019b; Marchiori, 2019c; Marchiori, 2019d; Marchiori, 2021b; Cingolani *et al.*, 2025).

Studies on parasitoids associated with synanthropic flies are essential for understanding ecological interactions and for supporting biological control programs. The identification of parasitoid species, their hosts, substrates, and parasitism rates provides important information for the management of dipteran populations of medical, sanitary, veterinary, and

forensic importance. In addition, studies conducted in different environments allow comparisons of parasitoid occurrence among rural, urban, forest, pasture, and livestock systems (Marchiori, 2019c; Cingolani *et al.*, 2025).

The experimental articles analyzed in the present synthesis provided quantitative data on pupae, parasitized pupae, parasitoid abundance, host species, substrates, and collection methods. These data made it possible to perform complementary chi-square analyses to test differences in parasitoid abundance, parasitism rates, host association, substrate preference, trap efficiency, and environmental variation. However, in several of the original articles, these statistical analyses had not been performed or presented. Therefore, the present study adds a new analytical contribution by applying chi-square tests to datasets that were previously interpreted mainly in descriptive form.

This study aimed to identify and synthesize parasitoid species associated with dipteran pupae collected in different substrates, evaluate their occurrence in relation to host species and environmental conditions, and contribute to the knowledge of parasitoid diversity associated with synanthropic flies. In addition, the study aimed to apply complementary chi-square analyses to available quantitative datasets, providing stronger statistical support for ecological patterns involving Diptera, parasitoids, substrates, habitats, and sampling methods, and generating information that may support future biological control programs targeting dipteran populations.

2.0. MATERIALS AND METHODS

2.1 General Approach

This study was conducted as a review of experimental works related to parasitoids associated with dipteran pupae. Scientific articles published in national and international journals were consulted, focusing on studies that evaluated parasitoid species, host associations, parasitism rates, and ecological interactions in different substrates. The selection of studies was based on their relevance to the topic, data availability, and methodological consistency. Only experimental studies involving the collection of dipteran pupae and the emergence of parasitoids were considered. Information regarding host species, parasitoid species, type of substrate, and parasitism rates was extracted and analyzed. Data obtained from the selected studies were organized and compared to identify patterns related to parasitoid diversity, host preference, and environmental influence. The results were interpreted considering ecological aspects and the potential application of parasitoids in biological control programs.

2.2. Experimental Methods

The study was carried out using animal feces as substrate for the collection of dipteran pupae and their associated parasitoids. Samples of fresh bovine and chicken feces were collected in rural environments and exposed under field conditions for a defined period to allow colonization by dipteran species. After exposure, the material was taken to the laboratory and placed in plastic containers containing a layer of sand as substrate for pupation. After approximately 15 days, the sand was sieved, and the pupae were recovered by flotation and individually placed in gelatin capsules until the emergence of adult flies or parasitoids. The emerged insects were identified based on morphological characters, and the association between host and parasitoid species was recorded (Figures 1-3).



Figure 1: Pastures: Artificial plate of bovine feces placed on soil within a container under field conditions. The figure illustrates the experimental setup used for attracting and collecting dipteran insects associated with decomposing organic matter. The yellow marker indicates the sampling point, facilitating identification and monitoring during the exposure period

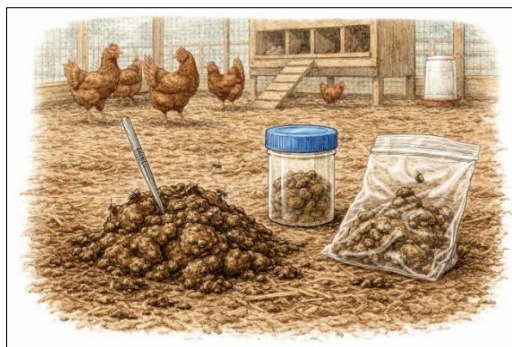


Figure 2: Chicken feces collected in a poultry house were used as substrate for insect sampling. The organic material provides suitable conditions for the development of dipteran larvae and their associated parasitoids. This method is commonly applied in studies of insect diversity and ecological interactions in poultry environments.



Figure 3: Fresh bovine feces deposited in a cattle corral, illustrating a substrate used for insect sampling. The organic material provides suitable conditions for colonization by coprophagous insects and their parasitoids this type of substrate is widely used in studies of insect succession and ecological interactions

Metal traps were constructed using cylindrical cans approximately 19 cm high and 9 cm in diameter, each containing two lateral rectangular openings in the lower portion to permit insect entry. At the top of each can, a nylon funnel open at both ends was attached, with the wider opening facing downward. Plastic bags were fitted to the upper part of the funnels to facilitate the collection of captured insects. Baits used to attract flies included bovine kidneys, human feces, chicken viscera,

fish, fruits, and bovine liver, all placed inside the cans over a layer of soil. Five traps were suspended from eucalyptus trees at a height of 1 m above ground level, spaced 2 m apart and located 50 m from domestic waste. The traps were suspended on *Eucalyptus* sp. trees at approximately 1 m above ground, spaced 2 m apart, and positioned about 50 m from areas containing domestic waste (Figure 4).



Figure 4: Adult flies were collected using traps constructed from matte, black-painted metal cans measuring approximately 19 cm in height and 9 cm in diameter. Each trap had two louver-type openings located in the lower third to allow insect entry. Nylon funnels, open at both ends, were attached to the top of the cans with their bases facing downward and connected to plastic bags, which were removed for fly collection

Pitfall traps were used to collect insects associated with dipteran pupae under field conditions. The traps consisted of plastic containers buried in the soil with their upper edges positioned at ground level, allowing the capture of ground-active insects. Each container was partially filled with a preservative solution composed of water, detergent, and a small amount of formalin or alcohol to reduce surface tension and preserve the collected specimens. In some cases, bait such as decomposing organic matter was added to increase attractiveness. The traps were distributed in the

study area with standardized spacing between them, depending on environmental conditions and sampling design. The exposure time varied according to the objectives of the study, generally ranging from 24 hours to several days. After the sampling period, the material collected in the traps was removed and taken to the laboratory for sorting. Insects were separated, counted, and preserved in 70% alcohol for identification. When dipteran pupae were present, they were isolated and maintained individually in gelatin capsules until the emergence of adult flies or parasitoids (Figure 5).



Figure 5: Pitfall trap used for the collection of ground-dwelling insects in field conditions. The trap consists of a container buried at soil level, allowing insects to fall and be retained. This method is widely used for sampling arthropods associated with soil and decomposing organic matter

The baits were exposed to environmental conditions in two distinct locations: a pasture area and a forest area, respectively, one under sunlight and the other in the absence of direct sunlight. The animals were euthanized mechanically by a blow to the head. Each bait was protected by a metal cage that allowed the entry of insects but prevented access by larger animals. The carcasses were placed on a metal plate fixed to the

ground, avoiding direct contact with the soil. Sawdust was placed on the plate and served as a substrate for the pupation of dipteran larvae. The pupae were collected by frequent sieving of the sawdust and were placed in glass containers until the emergence of adult flies and/or parasitoids. The adults obtained were counted and identified (Figure 6).



Figure 6: Baited trap using an animal carcass to attract necrophagous insects under field conditions. The decomposing organic matter serves as a resource for oviposition and development of dipteran species. This method is widely used to study insect succession and parasitoid–host interactions in decomposing substrates

2.3. Data Analysis

Parasitoid host preference was evaluated using Chi-square (χ^2) ($P < 0.05$) and analysis of variance (ANOVA) ($P < 0.0001$). Parasitism rate was calculated as: Number of parasitized pupae of each parasitoid species / total number of pupae of the host $\times 100$. The obtained data were organized according to substrate type,

host species, parasitoid species, number of individuals, number of parasitized pupae, and parasitism rate.

2.4. Note

Figures include both photographic records and schematic illustrations. Wing venation was omitted or simplified in schematic figures for clarity and does not

represent diagnostic morphological variation among parasitoid taxa.

3.0. RESULTS

The analysis of experimental studies revealed a wide diversity of parasitoid species associated with dipteran pupae collected in different substrates. The results showed variation in parasitoid occurrence according to host species, substrate type, and environmental conditions (Tables 1 and 2). The highest abundance of parasitoids was observed in substrates such as animal carcasses and organic waste, where greater availability of hosts favors parasitoid development. In

contrast, substrates such as feces and decomposing plant material showed lower parasitoid occurrence, indicating differences in ecological suitability.

The most frequently recorded parasitoid species included *Nasonia vitripennis* (Walker, 1836) (Hymenoptera: Pteromalidae), *Pachycrepoideus vindemmiae* (Rondani, 1875) (Hymenoptera: Pteromalidae), *Paraganaspis egeria* Diaz, Gallardo & Wash, 1996 (Hymenoptera: Pteromalidae), *Spalangia* spp. (Hymenoptera: Pteromalidae) and demonstrating their wide distribution and adaptability to different hosts and environments. The relative frequency of these species is presented in Table 1.

Table 1: Parasitoid species associated with dipteran pupae reported in experimental studies. Qualitative distribution of parasitoid species according to the literature. No numerical values are presented to avoid data inconsistency

Parasitoid species	Host association	Substrate	Biological type
<i>Brachymeria podagrica</i>	Diptera pupae	Carcass	Solitary
<i>Hemencyrtus herbertii</i>	Diptera pupae	Viscera	Gregarious
<i>Nasonia vitripennis</i>	Diptera pupae	Carcass	Gregarious
<i>Pachycrepoideus vindemmiae</i>	Diptera pupae	Carcass	Solitary
<i>Paraganaspis egeria</i>	Diptera larvae	Feces	Solitary
<i>Spalangia</i> spp.	Diptera pupae	Feces	Solitary
<i>Tachinobia</i> sp.	Diptera pupae	Organic matter	Gregarious
<i>Trichopria</i> sp.	Diptera pupae	Organic matter	Variable
Others	Various Diptera	Multiple substrates	Variable

Parasitism rates varied among studies and host species, reflecting differences in host density, substrate conditions, and parasitoid behavior. In general, higher parasitism levels were associated with environments that

provide favorable conditions for host development and parasitoid activity. These variations are summarized in Table 2.

Table 2: Occurrence of parasitoids according to substrate type in experimental studies. Qualitative comparison of substrates and parasitoid occurrence. No numerical values are presented to maintain scientific accuracy

Substrate	Host availability	Parasitoid occurrence	Ecological relevance
Animal viscera	High	High	Favorable
Bovine liver	Moderate	Moderate	Intermediate
Carcass	High	High	Major source
Chicken viscera	Moderate	Moderate	Intermediate
Fish	Low	Low	Occasional
Human feces	Moderate	Moderate	Relevant
Organic waste	High	High	Important
Plant material	Low	Low	Limited
Urban waste	Variable	Variable	Context dependent

The results indicate that parasitoid diversity and efficiency are influenced by multiple ecological factors, reinforcing the importance of environmental conditions and host availability in determining parasitism patterns.

4.0. EXPERIMENTAL MANUSCRIPTS

1. *Brachymeria Podagrica* Collected in Brazil

A total of 2,430 dipteran pupae were collected, from which 395 specimens of *B. podagrica* emerged, resulting in an overall parasitism rate of 16.3%. This relatively high parasitism rate may be associated with

host availability, host density, and the parasitoid's searching capacity. The species *B. podagrica* is widely distributed and is commonly associated with synanthropic Diptera emerging from pupae. This parasitoid was recorded developing in rat carcasses in tropical forest areas in the state of Goiás, Brazil, with a preference for *Pattonella intermutans* (Thomson, 1869) (Diptera: Sarcophagidae), from which predominantly female parasitoids emerged. The species has also been reported parasitizing pupae of *Sarcophaga* sp. (Diptera: Sarcophagidae) in decomposing carcasses (Table 3).

Table 3: Hosts of the parasitoid *Brachymeria podagrica* collected in the south of Goiás and west Minas Gerais, Brazil

Taxonomic group	Hosts species	Number of pupae	Number of pupae parasitized	Parasitism rate (%)
Calliphoridae	<i>Chrysomya albiceps</i>	20	9	45.0
	<i>Chrysomya megacephala</i>	19	1	5.3
	<i>Hemilucilia flavifacies</i>	43	3	7.0
Muscidae	<i>Ophyra</i> sp.	26	1	3.8
Sarcophagidae	<i>Oxysarcodexia thornax</i>	390	24	6.2
	<i>Peckia chrysostoma</i>	1324	245	18.5
	<i>Sarcodexia lambens</i>	608	112	18.4
Total		2430	395	16.3

In the present study, *C. albiceps* showed the highest of parasitism rate (45.0%), indicating that this host is highly suitable for the development of *B. podagrica*. This high parasitism rate may be influenced by factors such as resource quality, host availability, and methodological aspects. The species *C. albiceps* and *Chrysomya megacephala* (Fabricius, 1794) (Diptera: Calliphoridae) are of medical and sanitary importance, as they are associated with myiasis and the transmission of

pathogenic microorganisms. *Chrysomya megacephala* is commonly found in environments modified by human activity, where it develops in decomposing organic matter, including animal remains, and is frequently observed in urban areas. Its attraction to fermenting substances, decomposing materials, blood, and wounds increases its relevance in public health contexts (Figure 7).

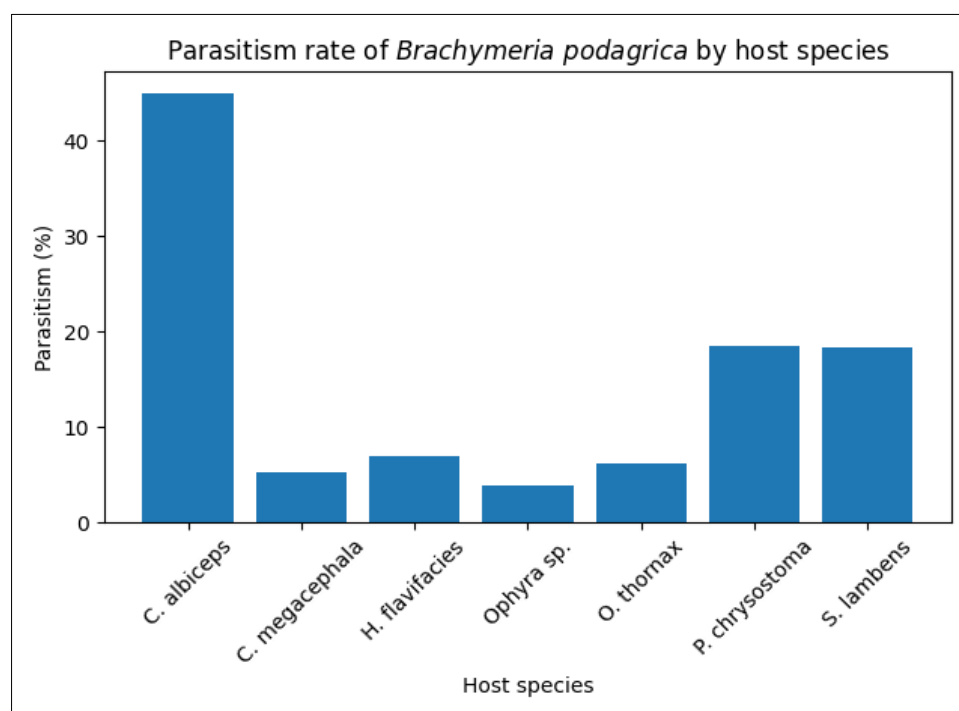


Figure 7: Parasitism rate of *Brachymeria podagrica* on dipteran hosts. Distribution of parasitism among different host species. Higher parasitism was observed in *Chrysomya albiceps* compared to other hosts

The occurrence of *B. podagrica* in association with these dipteran species contributes to the understanding of the bioecology and geographical distribution of parasitoids in Brazil. The use of insecticides for dipteran control often leads to the selection of resistant populations, reinforcing the need

for alternative control strategies. In this context, parasitoids such as *B. podagrica* may play an important role as natural enemies, contributing to the reduction of synanthropic Diptera populations and supporting biological control programs (Figure 8) (Marchiori, 2019a).



Figure 8: Adult of *Brachymeria podagrica* Synanthropic dipteran parasitoids, collected from different substrates in Itumbiara, Southern Goiás, Brazil. Source: <https://indiabiodiversity.org/observation/show/19194170>

2. *Hemencyrtus Herbertii* Collected in Brazil

A total of 1,336 dipteran pupae were collected, from which 31 specimens of *H. herbertii* emerged, resulting in an overall parasitism rate of 2.3%. Table 4 presents the hosts of *H. herbertii* and their respective parasitism rates. Among Calliphoridae, *C. albiceps* showed 3 parasitized pupae from 74 pupae (4.1%), *C. megacephala* had 1 parasitized pupa from 76 pupae (1.3%), *Hemilucilia flavifacies* Engel, 1931 (Diptera: Calliphoridae) had 3 parasitized pupae from 16 pupae (18.8%), and *Lucilia eximia* Wiedemann (1819) (Diptera: Calliphoridae) had 3 parasitized pupae from 268 pupae (1.1%).

Among Muscidae, *M. domestica* had 1 parasitized pupa from 20 pupae (5.0%), and *Ophyra* sp. (Diptera: Muscidae) presented 3 parasitized pupae from 64 pupae (4.7%). Among Sarcophagidae, *Euboettcheria collusor* Curran and Walley, 1934 (Diptera: Sarcophagidae) had 1 parasitized pupa from 412 pupae (0.2%), *Oxysarcodexia thornax* (Walker, 1849) (Diptera: Sarcophagidae) had 2 parasitized pupae from 109 pupae (1.8%), *Peckia chrysostoma* (Wiedemann, 1830) (Diptera: Sarcophagidae) had 9 parasitized pupae from 280 pupae (3.2%), and *S. lambens* showed the highest parasitism rate, with 5 parasitized pupae from 17 pupae (29.4%) (Table 4).

Table 4: Distribution of dipteran hosts of *Hemencyrtus herbertii* with number of pupae, parasitized pupae, and parasitism rate

Taxonomic group	Host species	Number of pupae	Parasitized pupae	Parasitism rate (%)
Calliphoridae	<i>Chrysomya albiceps</i>	74	3	4.1
	<i>Chrysomya megacephala</i>	76	1	1.3
	<i>Hemilucilia flavifacies</i>	16	3	18.8
	<i>Lucilia eximia</i>	268	3	1.1
Muscidae	<i>Musca domestica</i>	20	1	5.0
	<i>Ophyra</i> sp.	64	3	4.7
Sarcophagidae	<i>Euboettcheria collusor</i>	412	1	0.2
	<i>Oxysarcodexia thornax</i>	109	2	1.8
	<i>Peckia chrysostoma</i>	280	9	3.2
	<i>Sarcodexia lambens</i>	17	5	29.4
Total		1336	31	2.3

The results indicate that *H. herbertii* parasitized hosts from three dipteran families, demonstrating a relatively broad host range. However, parasitism rates varied considerably among host species, with *S. lambens* presenting the highest frequency. This variation may be related to differences in host availability, resource

quality, environmental conditions, and parasitoid searching behavior. The occurrence of higher parasitism in certain hosts suggests that host suitability and density may influence parasitoid efficiency. In addition, biological characteristics of parasitoids may affect their success in exploiting specific hosts (Figures 9-10).

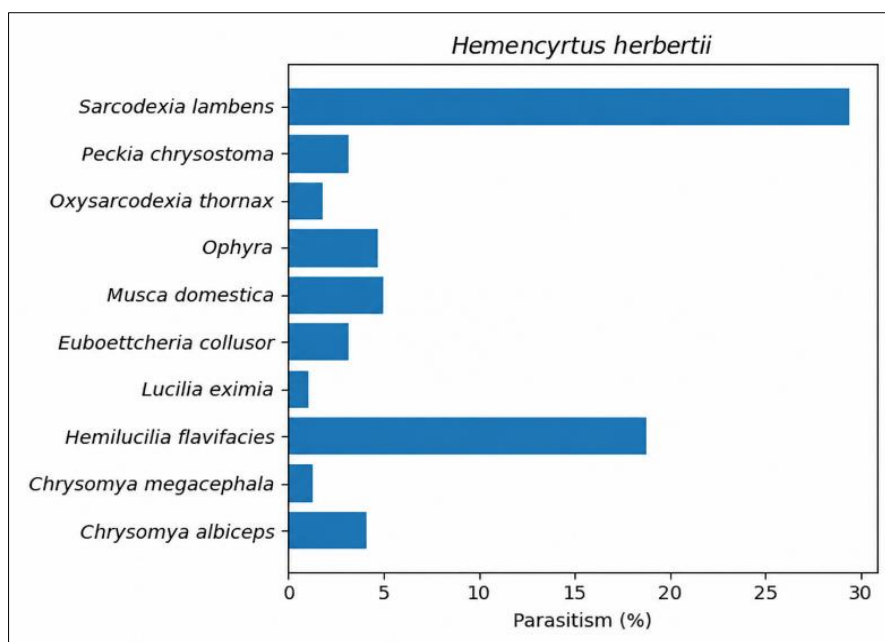


Figure 9: Parasitism rate of *Hemencyrtus herbertii* on dipteran hosts. The distribution of parasitism among host species shows variation in host susceptibility, with the highest parasitism rate recorded in *Sarcodexia lambens* compared with the other hosts



Figure 10: Males of Sarcophagidae (Diptera) species of possible forensic interest found in rabbit carcasses in Curitiba, Paraná. Source: Vairo, K. P. (2008). Federal University of Paraná

Among the collected hosts, *C. albiceps*, *C. megacephala*, and *M. domestica* stand out for their medical and veterinary importance, as they are associated with pathogen transmission and myiasis. The chi-square analysis showed a significant difference in parasitism among dipteran host species ($\chi^2 = 88.29$; $df = 9$; $p < 0.0001$), indicating that parasitism by *H. herbertii* was not evenly distributed among hosts. The presence of *H. herbertii* parasitizing species of sanitary importance highlights its potential role as a natural enemy of synanthropic Diptera. These results reinforce the importance of parasitoids in regulating fly populations and suggest that *H. herbertii* may contribute to the natural control of flies in environments rich in decomposing organic matter, such as human feces and animal remains (Marchiori, 2014a; Marchiori, 2014b; Marchiori, 2019b).

3. Parasitoids of *Musca Domestica* Collected in two Areas of the Central Region of Goiás, Brazil

Parasitoids of *M. domestica* were collected in rural and urban areas of the central region of Goiás, Brazil. A total of 822 pupae of *M. domestica* were examined. In the rural area, 350 pupae were collected, of which 24 were parasitized, resulting in a parasitism rate of 6.9%. In the urban area, 472 pupae were collected, of which 42 were parasitized, corresponding to a parasitism rate of 8.9%. Considering all parasitoids obtained, 24 individuals (36.4%) were recorded in the rural area and 42 individuals (63.6%) in the urban area. Table 5 presents the parasitoid species associated with *M. domestica* and their respective parasitism rates in both environments.

Table 5: Frequency of parasitism of *Musca domestica* collected in two areas of the central region of Goiás, Brazil. Distribution of parasitoid species in rural and urban areas, with the number of parasitized pupae and the parasitism rate

Taxonomic Group	Parasitoid species	Rural		Urban	
		Number of pupae parasitized	Parasitism rate (%)	Number of pupae parasitized	Parasitism rate (%)
Encyrtidae	<i>Hemencyrtus herbertii</i>	5	1.4	1	0.2
Eulophidae	<i>Tachinobia</i> sp.	–	–	1	0.2
Pteromalidae	<i>Nasonia vitripennis</i>	–	–	7	1.5
	<i>Pachycrepoideus vindemmiae</i>	19	5.4	26	5.5
	<i>Spalangia</i> sp.	–	–	7	1.5
Total of pupae parasitized	–	24	6.9	42	8.9
Total	–	350	-	472	-

Among the parasitoids recorded, *P. vindemmiae* was the most frequent species in both areas, with 19 parasitized pupae (5.4%) in the rural area and 26 parasitized pupae (5.5%) in the urban area. This species showed the highest contribution to parasitism, indicating adaptation to both environments and efficiency in exploiting *M. domestica* pupae. Other parasitoids were recorded with lower frequencies. In the rural area, *H*

herbertii was responsible for 5 parasitized pupae (1.4%). In the urban area, *H. herbertii*, *Tachinobia* sp. (Hymenoptera: Eulophidae), *N. vitripennis*, and *Spalangia* sp. were recorded, with 1, 1, 7, and 7 parasitized pupae, respectively. These species contributed to parasitoid diversity, although with lower parasitism rates (Figure 11).

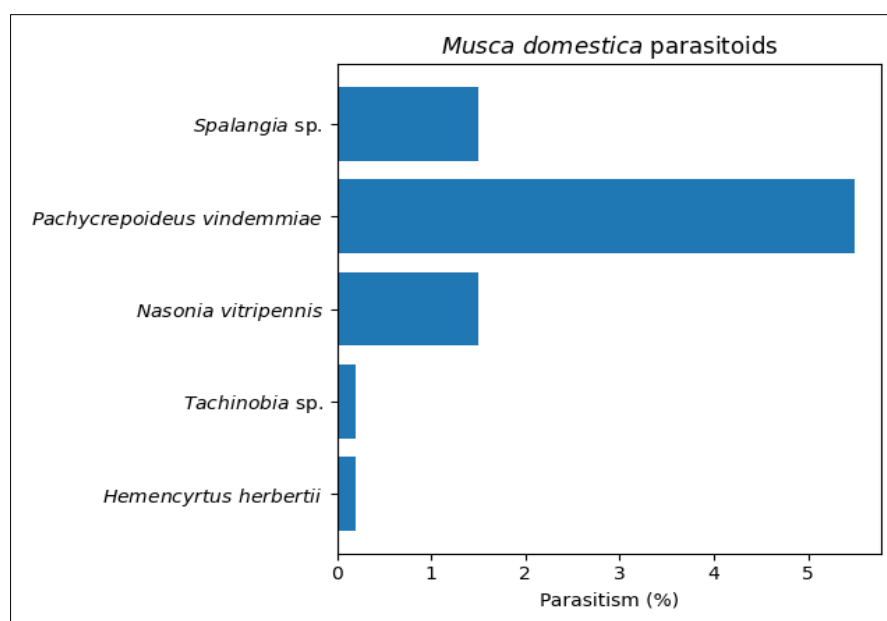


Figure 11: Parasitism of parasitoids associated with *Musca domestica* in rural and urban environments. Distribution of parasitoid species with emphasis on *Pachycrepoideus vindemmiae*, the most frequent species in both areas

The comparison between environments showed that the overall parasitism rate was numerically higher in the urban area than in the rural area. However, the chi-square test comparing parasitized and non-parasitized pupae between environments did not show a significant difference ($\chi^2 = 1.13$; $df = 1$; $p = 0.287$), indicating that the overall parasitism rate was statistically similar between the two areas. In contrast, the distribution of

parasitoid species differed significantly between rural and urban environments ($\chi^2 = 14.96$; $df = 4$; $p = 0.0048$), with greater parasitoid diversity recorded in the urban area. This result suggests that urban environments may provide a wider range of organic substrates and microhabitats suitable for different parasitoid species (Figure 12).

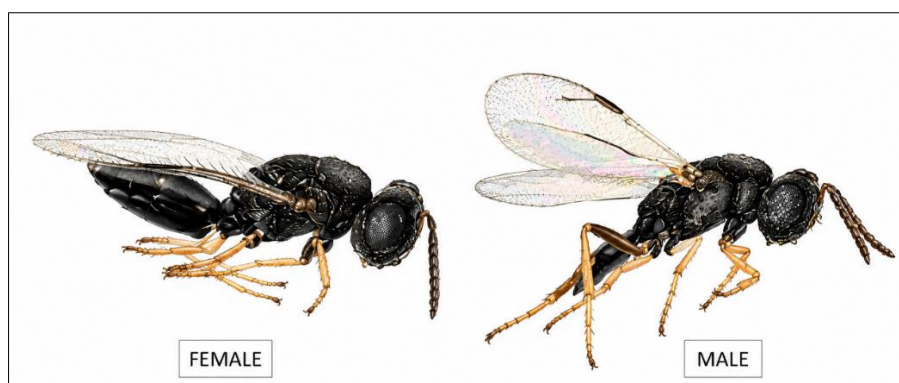


Figure 12: Adult of *Pachycrepoideus vindemmiae*. Use of an insecticide can be counterproductive if it also harms natural enemies of a target pest. A new study puts several insecticides currently in use to manage spotted-wing *Drosophilidae* to the test to see how they do or don't affect parasitoids of the invasive fruit fly. Source: Entomological Society of America, Annapolis, Maryland, USA

The predominance of *P. vindemmiae* may be explained by its wide host range, highly competitive ability, and broad geographic distribution. This species is known to parasitize several Diptera families and is frequently associated with synanthropic flies. The presence of parasitoids such as *H. herbertii*, *N. vitripennis*, *Spalangia* sp., and *Tachinobia* sp. reinforces the ecological importance of natural enemies in regulating *M. domestica* populations. These results suggest that both rural and urban areas may support parasitoid communities capable of contributing to the natural control of flies of sanitary importance, especially in environments rich in organic matter (Marchiori, 2014a; Marchiori, 2014b; Marchiori, 2019b).

4. Parasitoids of Dipterans Collected from Animal Excrement in Southern Goiás

A total of 77 parasitoid specimens were obtained from 628 dipteran pupae collected in bovine feces, whereas 310 parasitoids were obtained from 3,116 pupae collected in chicken feces. Thus, chicken manure

produced a higher absolute number of parasitoids, while the overall parasitism rates were 12.3% in bovine feces and 10.0% in chicken feces. The comparison of overall parasitism between substrates was not statistically significant ($\chi^2 = 3.02$; $df = 1$; $p = 0.082$), indicating that the difference was mainly related to the larger number of pupae obtained from chicken feces.

In bovine feces, the most frequent parasitoid species was *Spalangia cameroni* Perkins 1910 (Hymenoptera: Pteromalidae), with 26 parasitized pupae. The highest host-level parasitism rate was observed in *O. thornax*, with 20 parasitized pupae among 70 collected, corresponding to 28.6%. Other relevant host-level parasitism rates were recorded in *R. belforti*, with 16 parasitized pupae from 63 pupae (25.4%), *B. quadristigma*, with 10 parasitized pupae from 49 pupae (20.4%), and *Archiseopsis scabra* (Loew 1861) (Diptera: Sepsidae), with 4 parasitized pupae from 40 pupae (10.0%) (Table 6).

Table 6: Parasitoids of synanthropic dipterans collected in cattle dung in Itumbiara, Goiás, Brazil. Distribution by host species and parasitoid species, with the number of parasitized pupae and frequency of parasitism

Host species	No. individuals	Parasitoid species	No. parasitized	Parasitism rate (%)
<i>Archiseopsis scabra</i>	40	<i>Spalangia drosophilae</i>	4	10.0
<i>Brontaea debilis</i>	56	<i>Spalangia cameroni</i>	1	1.8
		<i>Spalangia nigroaenea</i>	2	3.6
<i>Brontaea quadristigma</i>	49	<i>Kleidotoma nigra</i>	2	4.1
		<i>Spalangia cameroni</i>	1	2.0
		<i>Spalangia drosophilae</i>	1	2.0
		<i>Spalangia endius</i>	1	2.0
		<i>Spalangia nigroaenea</i>	5	10.2
		<i>Spalangia nigra</i>	3	2.0
<i>Cyrtoneurina paraescita</i>	151	<i>Spalangia nigroaenea</i>	5	3.3
<i>Musca domestica</i>	10	Absent	0	0.0
<i>Oxysarcodexia thornax</i>	70	<i>Spalangia cameroni</i>	20	28.6
<i>Palaeosepsis</i> spp.	107	<i>Gnathopleura quadridentata</i>	2	1.9
		<i>Paraganaspis egeria</i>	2	1.9
		<i>Spalangia drosophilae</i>	1	0.9
		<i>Spalangia endius</i>	2	1.9

		<i>Triplasta atrocotalis</i>	8	7.5
		<i>Triplasta cotalis</i>	1	0.9
<i>Ravinia belforti</i>	63	<i>Pachycrepoideus vindemmiae</i>	1	1.6
		<i>Spalangia cameroni</i>	4	6.3
		<i>Spalangia nigra</i>	6	9.5
		<i>Trichopria</i> sp.	5	7.9
Other species not parasitized	82			
Total	628		77	12.3

In chicken feces, *Spalangia endius* Walker, 1839 (Hymenoptera: Pteromalidae) was the dominant species, with 147 parasitized pupae, mainly associated with *M. domestica*. *P. vindemmiae* was also an important component of the parasitoid community, with 65

parasitized pupae, followed by *S. cameroni*, with 38 parasitized pupae. These results indicate that chicken feces supported a higher abundance and greater diversity of parasitoids, mainly associated with *M. domestica* (Figures 13–14) (Table 7).



Figure 13: Parasitoids associated with dipterans collected in bovine dung. *Spalangia cameroni* was the predominant parasitoid, mainly associated with *Oxysarcodexia thornax*. Source: <https://en.wikipedia.org/wiki/Spalangia>

Table 7: Parasitoids of synanthropic dipterans collected in chicken feces in Itumbiara, Goiás, Brazil. Distribution by host species and parasitoid species, with the number of parasitized pupae and frequency of parasitism

Hosts species	No. individuals	Parasitoid species	No. parasitized	Parasitism rate (%)
<i>Fannia pusio</i>	153	<i>Eurytoma</i> sp.	2	1.3
		<i>Pachycrepoideus vindemmiae</i>	9	5.9
<i>Musca domestica</i>	1982	<i>Aphaereta</i> sp.	1	0.1
		<i>Nasonia vitripennis</i>	11	0.6
		<i>Pachycrepoideus vindemmiae</i>	52	2.6
		<i>Spalangia cameroni</i>	38	1.9
		<i>Spalangia drosophilae</i>	1	0.1
		<i>Spalangia endius</i>	143	7.2
		<i>Spalangia nigra</i>	3	0.2
		<i>Spalangia nigroaenea</i>	10	0.5
		<i>Spalangia</i> sp.	21	1.1
		<i>Tachinaephagus zealandicus</i>	1	0.1
<i>Ophyra</i> sp.	529	<i>Nasonia vitripennis</i>	1	0.2
		<i>Spalangia endius</i>	4	0.8
<i>Ornidia obesa</i>	226	<i>Pachycrepoideus vindemmiae</i>	2	0.9
<i>Sarcodexia lambens</i>	222	<i>Pachycrepoideus vindemmiae</i>	2	0.9
		<i>Tachinaephagus zealandicus</i>	9	4.1
Other species not parasitized	4			
Total	3116		310	10.0



Figure 14: Parasitoids associated with dipterans collected in chicken feces. *Spalangia endius* was the predominant species, especially in *Musca domestica* pupae. Source: <https://bugsforbugs.com.au/product/spalangia/>

Host–parasitoid associations differed significantly in bovine feces ($\chi^2 = 229.24$; $df = 66$; $p < 0.0001$). According to the data, *S. cameroni* was associated with *Brontaea debilis* (Williston, 1896) (Diptera: Muscidae), *B. quadristigma*, *O. thornax*, and *R. belforti*; *Spalangia nigroaenea* Curtis, 1839 (Hymenoptera: Pteromalidae) was associated with *B. debilis*, *B. quadristigma*, and *Cyrtoneurina paraescita* (Couri, 1995) (Diptera: Muscidae); *Spalangia drosophilae* Ashmead, 1885 (Hymenoptera: Pteromalidae) was associated with *A. scabra*, *B. quadristigma*, and *Palaeosepsis* sp.; *S. endius* was associated with *B. quadristigma* and *Palaeosepsis* sp.; *Spalangia nigra* Latreille 1805 (Hymenoptera: Pteromalidae) was associated with *C. paraescita* and *R. belforti*; and other parasitoids, including *G. quadridentata*, *Kleidotoma nigra* (Hartig, 1840) (Hymenoptera: Figitidae), *P. egeria*, *Trichopria* sp., *Triplasta atrocoxalis* (Ashmead, 1895) (Hymenoptera: Figitidae), *Triplasta coxalis* (Ashmead, 1895) (Hymenoptera: Figitidae), and *P. vindemmiae*, occurred in specific host associations.

In chicken feces, host–parasitoid associations were also significant ($\chi^2 = 325.17$; $df = 40$; $p < 0.0001$). *Aphaereta* sp. and *Eurytoma* sp. (Hymenoptera: Eurytomidae) were associated with *M. domestica* and *Fannia pusio* (Wiedemann, 1830) (Diptera: Fanniidae), respectively; *N. vitripennis* was associated with *M.*

domestica and *Ophyra* sp.; *P. vindemmiae* was associated with *F. pusio*, *M. domestica*, *Ornidia obesa* (Fabricius, 1775) (Diptera: Syrphidae), and *S. lambens*; *S. cameroni*, *S. drosophilae*, *S. endius*, *S. nigra*, *S. nigroaenea*, and *Spalangia* sp. were associated mainly with *M. domestica*; and *Tachinaephagus zealandicus* Ashmead, 1904 (Hymenoptera: Encyrtidae) was associated with *M. domestica* and *S. lambens*. These results show that substrate type and host availability influenced parasitoid occurrence and host use in animal excrement (Figure 12) (Marchiori *et al.*, 2010).

5. Techniques for Collecting and Capturing Insects of the Orders Diptera and Hymenoptera Collected in Goiás

Four trap types were evaluated for collecting Diptera and Hymenoptera in the state of Goiás: Moericke traps, Malaise traps, metallic container traps, and pitfall traps. These methods differ in structure, efficiency, selectivity, and applicability according to the target insect group and environmental conditions. Moericke traps are yellow containers filled with a liquid solution that visually attracts insects. This method is inexpensive, easy to use, and useful for sampling parasitoid Hymenoptera in natural and cultivated environments. However, its efficiency may vary according to climatic conditions, and periodic maintenance is required because of evaporation and contamination of the solution (Figure 15).

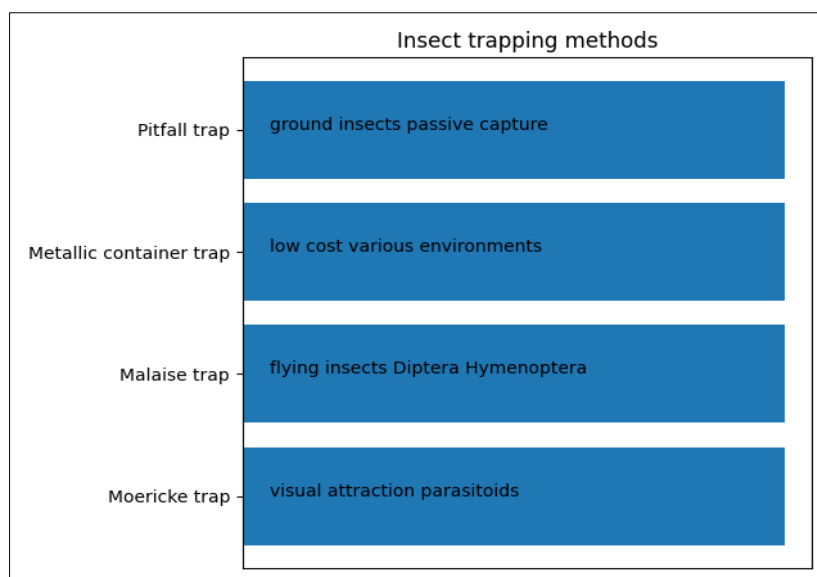


Figure 15: Trap models used for collecting Diptera and Hymenoptera in Goiás, Brazil, including Moericke traps, Malaise traps, metallic container traps, and pitfall traps

Malaise traps collect flying insects by interception, directing specimens into collecting containers. They are effective for sampling Diptera and Hymenoptera, especially in forested environments. However, their efficiency is mainly restricted to flying insects and may be reduced in open areas. Metallic container traps are low-cost and easy to construct, and they can be used in forest, rural, and urban environments. Nevertheless, the lack of standardization among models may reduce comparability among studies, and

environmental factors such as rainfall and temperature can influence trap performance.

Pitfall traps are passive sampling devices designed to capture insects that move on the soil surface. They are simple, inexpensive, and widely used in ecological studies. These traps are particularly useful for ground-dwelling insects, but they may underestimate flying species and can be affected by rain, soil disturbance, and substrate conditions (Figure 16).

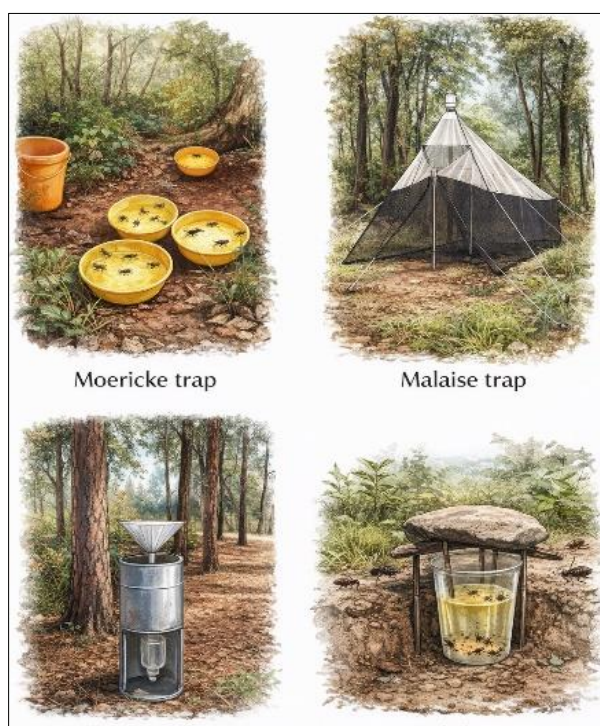


Figure 16: Insect trapping methods used for collecting Diptera and Hymenoptera. The illustration shows Moericke, Malaise, metallic containers, and pitfall traps in natural environments, emphasizing their different structures and sampling strategies according to insect behavior

Overall, each trap type presented specific advantages and limitations, indicating that no single method is sufficient to sample the full diversity of Diptera and Hymenoptera. The combined use of different trapping techniques is recommended to obtain more complete and representative samples of insect communities. Because the available information for this section is descriptive and does not provide quantitative abundance data for each trap type, chi-square analysis was not applied (Marchiori, 2016).

6. New Host and Superparasitism for *Spalangia drosophilae* in Brazil

A total of 27 pupae of *O. thornax* were collected using traps baited with bovine liver in a Cerrado area of Caldas Novas, Goiás, Brazil. From this 1 pupa, 23 specimens of *S. drosophilae* emerged, resulting in a parasitism rate of 3.7%. This study represents the first record of *S. drosophilae* parasitizing *O. thornax* in Brazil. The association expands the known host range of this parasitoid species, which is commonly reported to attack pupae of Diptera from several families, including Muscidae, Calliphoridae, Sarcophagidae, Drosophilidae, and others (Figure 17).



Figure 17: Superparasitism in dipteran pupae, showing multiple larvae developing within a single host. The image illustrates an open pupa containing numerous immature stages and a parasitized pupa with emerging adults. This pattern reflects the gregarious development and high parasitic pressure typical of *Spalangia drosophilae*

Superparasitism was observed, with up to 26 individuals emerging from a single host pupa. This behavior contrasts with previous reports suggesting that *S. drosophilae* tend to avoid superparasitism. The occurrence of multiple individuals in a single pupa is likely related to the larger size of *O. thornax* pupae, which may provide sufficient resources to support the development of more than one parasitoid.

Previous studies in Goiás reported *S. drosophilae* parasitizing other hosts, such as *A. scabra* and *Palaeosepsis* sp., with lower parasitism rate ranging from 0.8% to 5.7%. In comparison, the parasitism rate observed in *O. thornax* is a lower level of host suitability or greater efficiency of the parasitoid under the studied conditions. These results demonstrate that *S. drosophilae* is capable of exploiting new hosts and may exhibit flexible parasitic behavior depending on host

characteristics. The occurrence of superparasitism suggests adaptive strategies related to host size and resource availability, reinforcing the ecological importance of this species as a natural enemy of synanthropic flies (Marchiori *et al.*, 2006).

7. First Report of the Parasitoid *Paraganaspis Egeria* Parasitizing Immature Stages of *Fannia Pusio* in Brazil

A total of 99 pupae of *F. pusio* were collected from human feces in Itumbiara, Goiás, Brazil. From these pupae, three specimens of *P. egeria* emerged, resulting in a parasitism rate of 3.0%. This study represents the first record of *P. egeria* parasitizing immature stages of *F. pusio* in Brazil. This finding expands the known host range of this parasitoid, which had previously been reported parasitizing species such as *S. occidua*, *C. albiceps*, and *B. quadristigma* (Figure 18).



Figure 18: New host record of *Paraganaspis egeria* associated with *Fannia pusio*. Illustration of parasitoid development showing immature stages and emergence from the host pupa. Representation of host expansion and parasitic interaction in synanthropic environments

Species of Eucolilinae are endoparasitoids that oviposit in larval stages of Diptera and complete development within the host, emerging from the pupal stage. The relatively low parasitism rate observed 3.0% suggests limited efficiency or low host–parasitoid encounter frequency under the studied conditions. The occurrence of *P. egeria* in human feces indicates its adaptation to synanthropic environments, where hosts such as *F. pusio* develop in decomposing organic matter. This ecological association highlights the importance of parasitoids in regulating populations of flies of sanitary importance.

Overall, the results demonstrate that *P. egeria* is capable of exploiting new hosts in Brazil, reinforcing its

ecological role as a natural enemy of dipterans and contributing to the understanding of parasitoid diversity associated with synanthropic flies (Marchiori *et al.*, 2002a).

8. Parasitoids Hymenoptera Collected Using Different Trap Types in Brazil

A total of 400 parasitoids were collected using two trap types in southern Goiás, Brazil. Metal container traps captured 180 individuals, whereas pitfall traps captured 220 individuals, indicating a numerically higher abundance in pitfall traps. The overall frequencies were 45.0% for metal container traps and 55.0% for pitfall traps (Table 8).

Table 8: Parasitoids of dipterans collected using metal containers and pitfall traps in southern Goiás, Brazil. Distribution of parasitoid species according to trap type, with the number of individuals collected and relative frequency within each trap type

Taxonomic groups	Parasitoids species	Metal container traps	Frequency (%)	Pitfall traps	Frequency (%)
Braconidae	<i>Gnathopleura quadridentata</i>	22	12.2	157	71.4
Chalcididae	<i>Brachymeria podagrica</i>	21	11.7	0	
Encyrtidae	<i>Hemencyrtus herbertii</i>	23	12.8	0	
Figitidae	<i>Neralsia</i> sp.	0	-	43	19.5
	<i>Paraganaspis egeria</i>	1	0.6	0	
	<i>Triplasta</i> sp.	0	-	17	7.7
Pteromalidae	<i>Nasonia vitripennis</i>	13	7.2	0	-
	<i>Pachycrepoideus vindemmiae</i>	100	55.6	0	-
	<i>Spalangia endius</i>	0	-	2	0.9
	<i>Spalangia nigroaenea</i>	0	-	1	0.5
Total		180	100	220	100

The original analysis of variance reported no significant differences in trap attraction in relation to parasitoid species ($F = 0.4$; $P = 0.85$) or among parasitoid species ($F = 0.93$; $P = 0.54$). However, complementary chi-square analysis based on the absolute number of parasitoids showed a significant difference in total abundance between trap types ($\chi^2 = 4.00$; $df = 1$; $p =$

0.0455), with pitfall traps collecting more individuals. In addition, the distribution of parasitoid species differed significantly between trap types ($\chi^2 = 322.04$; $df = 9$; $p < 0.0001$), indicating that each trap type sampled different components of the parasitoid community.

In pitfall traps, the most frequent species was *G. quadridentata*, with 157 individuals, representing 71.4% of the parasitoids collected with this method. This high frequency may be related to its searching ability, seasonal occurrence, host availability, or competitive advantage during larval development. This species behaves as a solitary parasitoid, emerging as a single individual from each host pupa, and is known to parasitize Diptera of the families Sarcophagidae and Muscidae.

In metal container traps, the most frequent species was *P. vindemmiae*, with 100 individuals, corresponding to 55.6% of the parasitoids collected with this trap type. This predominance is likely associated with its polyphagous behavior and wide host range, which includes several Diptera families such as Calliphoridae, Muscidae, Sarcophagidae, and Tephritidae. Other species were recorded with lower frequencies, including *B. podagrica*, *H. herbertii*, *N. vitripennis*, *Neralsia* sp., *P. egeria*, *S. endius*, *S. nigroaenea*, and *Triplasta* sp., demonstrating the

diversity of parasitoids associated with dipteran hosts collected by different trapping methods.

The results indicate that trap type influenced both parasitoid abundance and species composition. Pitfall traps, as passive sampling methods, depend on insect activity and may capture a broader range of ground-associated species. In contrast, metal container traps are influenced by bait type and environmental conditions, which may affect capture efficiency and specimen preservation. Therefore, the significant chi-square result suggests that the two trap models were complementary rather than equivalent in the parasitoid assemblages sampled. Overall, the study demonstrates that both trapping methods are useful for collecting parasitoids of dipteran pupae, but their efficiency varies according to parasitoid species and sampling strategy. The combined use of metal containers and pitfall traps is recommended to obtain a more comprehensive representation of parasitoid diversity in different environments (Figure 19) (Marchiori, 2019d).



Figure 19: Trapping methods used for collecting parasitoids of dipterans in Brazil. The illustration shows metal containers and pitfall traps as complementary sampling strategies for parasitoid capture in natural environments. Differences in trap structure and field arrangement may influence parasitoid abundance and species composition

9. New Habitat and Host in *Trichopria* Sp. In Brazil

A total of 155 pupae of *P. chrysostoma* were obtained from traps baited with bovine kidney in Lavras, Minas Gerais, Brazil. From this 1 pupa, 41 specimens of *Trichopria* sp. emerged, resulting in a parasitism rate of 0.65%. Superparasitism was observed, with multiple individuals of *Trichopria* sp. emerging from a single host pupa. This behavior is characteristic of some parasitoids and may occur when several eggs are deposited in the same host, allowing the development of more than one individual within a single puparium.

Previous studies have reported *Trichopria* sp. parasitizing hosts such as *B. quadristigma*, *Coproica* sp. (Diptera: Sphaeroceridae), *Haematobia irritans* Linnaeus, 1758 (Diptera: Muscidae), *Palaeosepsis* spp., and *S. occidua* in bovine feces, generally behaving as a solitary parasitoid. In contrast, the present study demonstrated superparasitism in *P. chrysostoma*, indicating that parasitic behavior may vary depending on host characteristics and environmental conditions (Figure 20).

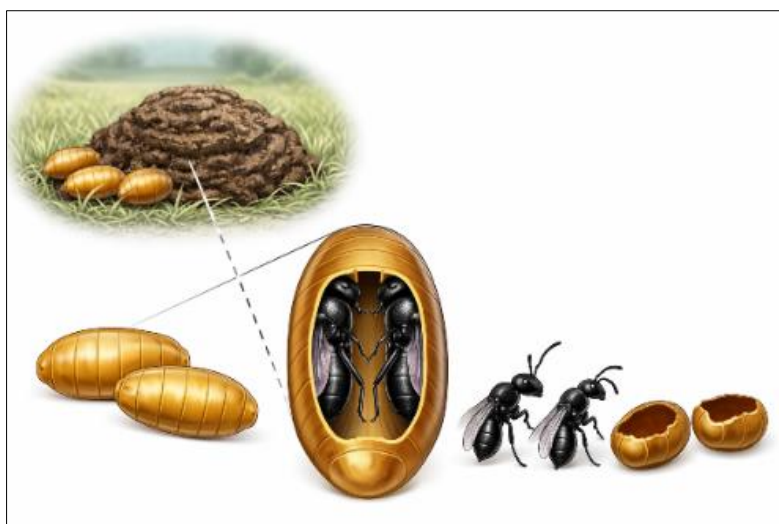


Figure 20: Superparasitism of *Trichopria* sp. in pupae of *Peckia chrysostoma*. Illustration showing the emergence of a single parasitoid individual from one host pupa. Representation of the solitary parasitism associated with this host–parasitoid interaction

The low parasitism rate observed may be associated with variations in resource availability, host density, or environmental factors affecting parasitoid efficiency. Despite this, the occurrence of *Trichopria* sp. in bovine kidney represents a new habitat record, expanding the ecological knowledge of this parasitoid species. These results demonstrate that *Trichopria* sp. can exploit new hosts and habitats, reinforcing its ecological importance as a natural enemy of dipterans and contributing to the understanding of parasitoid diversity in synanthropic environments (Marchiori, 2004a).

10. Parasitoid of Muscoid Dipterans Collected in Itumbiara, Goiás, Brazil

A total of 511 dipteran pupae were collected, of which 18 were parasitized by *H. herbertii*, producing 121 parasitoid individuals and resulting in an overall parasitism rate of 3.5%. The parasitoids were obtained from three substrates: bovine viscera, human feces, and bovine liver. The highest number of *H. herbertii* individuals was observed in bovine viscera (75 individuals), followed by human feces (23) and bovine liver (23) (Figure 21).

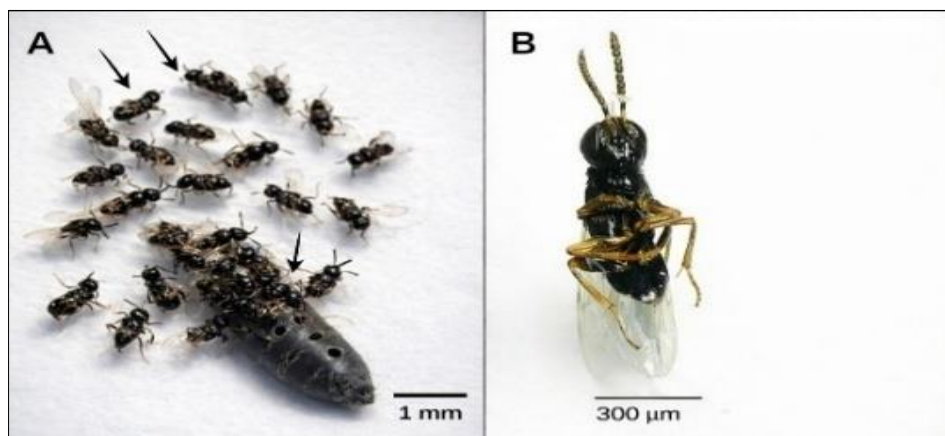


Figure 21: Gregarious emergence of *Hemencyrtus herbertii* from a dipteran host pupa. The figure illustrates multiple parasitoid individuals emerging from the same host puparium and an adult specimen in lateral view

Hemencyrtus herbertii was associated with several dipteran hosts distributed among different substrates. In bovine viscera, it parasitized *H. flavifacies* and *P. chrysostoma*. In human feces, the parasitism rate was recorded in *M. domestica* and *O. thornax*. In bovine liver, the hosts were *E. collusor* and *S. nudiseta*, demonstrating a broad host range.

Parasitism rate varied among host species, with the highest value observed in *H. flavifacies* (18.8%), followed by *M. domestica* (5.0%), *Synthesiomyia nudiseta* Van Der Wulp, 1883 (Diptera: Muscidae) (3.6%), *P. chrysostoma* (3.2%), *E. collusor* (3.2%), and *O. thornax* (1.8%). These results indicate differences in host suitability and parasitoid occurrence among the host species evaluated. The chi-square analysis showed a significant difference in parasitism rate among dipteran

host species (chi-square = 12.05; df = 5; p = 0.0341), indicating that parasitism rate by *H. herbertii* was not

evenly distributed among hosts, with a higher parasitism rate observed in *H. flavifacies* (Figure 22).



Figure 22: Gregarious parasitism rate of *Hemencyrtus herbertii* in dipteran pupae. The illustration shows multiple parasitoid individuals developing in the same host puparium, emphasizing parasitoid aggregation and emergence from a single parasitized pupa

According to the data presented in Table 9, the number of parasitoid individuals and parasitized pupae varied among substrates and hosts, reinforcing the influence of environmental conditions, host availability,

and parasitoid behavior on parasitism rate patterns. Because *H. herbertii* is a gregarious parasitoid, the number of emerged individuals was higher than the number of parasitized pupae.

Table 9: *Hemencyrtus herbertii* collected from dipteran pupae in different substrates in Itumbiara, Goiás, Brazil. Distribution of hosts, number of pupae, parasitoid individuals, parasitized pupae, and parasitism rate

Substrate	Hosts species	No. individuals	<i>H. herbertii</i>	Parasitized pupae	Parasitism rate (%)
Bovine liver	<i>Euboettcheria collusor</i>	31	3	1	3.2
	<i>Synthesiomys nudiseta</i>	55	20	2	3.6
Bovine viscera	<i>Hemilucilia flavifacies</i>	16	14	3	18.8
	<i>Peckia chrysostoma</i>	280	61	9	3.2
Human feces	<i>Musca domestica</i>	20	5	1	5.0
	<i>Oxysarcodexia thornax</i>	109	18	2	1.8
Total		511	121	18	3.5

Hemencyrtus herbertii behaves as a gregarious parasitoid, with multiple individuals developing within a single host and emerging from the pupal stage. This biological characteristic, combined with its ability to

parasitize different host species and occur in different decomposing substrates, highlights its ecological importance and potential role in the biological control of synanthropic flies (Figure 23) (Marchiori *et al.*, 2004).

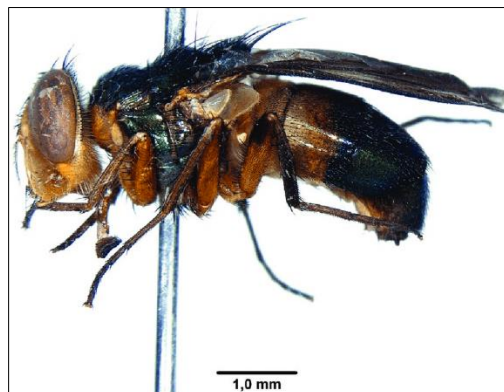


Figure 23: Parasitism rate by *H. herbertii* was not evenly distributed among hosts, with a higher parasitism rate observed in *H. flavifacies*. Ecological succession of blow flies (Diptera: Calliphoridae) in the decomposition of pig carcasses in Cerrado areas of Brazil. Source: Doi:10.1093/jme/tjad136

11. A New Species of *Steleucoela* N. Sp. From Brazil (Hymenoptera: Figitidae)

The genus *Steleucoela* is revised, and a new species, *Steleucoela brasiliensis*, is described based on specimens collected in Brazil. The material examined included seven individuals (two males and five females) obtained using Malaise traps in a remnant of native vegetation, expanding the known distribution of the genus to the Neotropical region. The diagnostic characteristics of the genus were updated, including features such as a wedge-shaped head in lateral view, a prominent ocellar tubercle, projected antennal sockets, and the presence of malar grooves. Additional characters include a sessile metasoma with a hairy ring at the base and forewings with a partially open radial cell, and

confirming previous revisions and clarifying morphological interpretation.

Steleucoela brasiliensis n. sp. is characterized by a smooth and polished body, weak pilosity, and a laterally compressed mesosoma. The head is wider than the mesosoma and wedge-shaped in lateral view, with cylindrical antennal segments progressively shortening toward the apex. The mesosoma is elongate, with pronotal sides entirely smooth and a well-defined scutellum with a posteriorly rounded margin. The propodeum presents distinct carinae, and the forewing radial cell is more than twice as long as wide (Figures 24A-24B).



Figure 24A: Morphological characters of *Steleucoela brasiliensis* n. sp. Female: head, lateral view; mesosoma, dorsal view; antenna; pronotal plate; mesoscutum and scutellum, dorsal view; scutellum and propodeum, dorsal view; and scutellum, lateral view. Male: antenna, segments 1–3; and radial cell of fore wing. Adapted as an original scientific plate based on Gallardo, Díaz, and Marchiori (2003)



Figure 24B: Gallardo, F. E., Díaz, N. B., & Marchiori, C. H. (2003). A new species of *Steleucoela* from Brazil (Hymenoptera: Figitidae). *Transactions of the American Entomological Society*, 129(1), 107–110. Source: <https://bioone.org/>

Sexual dimorphism was observed, with males being smaller (2.4 mm) than females (3.1 mm) and presenting 15-segmented antennae, while females have 13-segmented antennae. Variation among specimens included differences in body coloration and slight morphological variation in mesosoma and metasoma structures.

The new species differs from *Steleucoela piriformis* Kiefer, 1908 (Hymenoptera: Figitidae) mainly in the morphology of the head and mesosoma. In *S. brasiliensis*, the head is narrower in lateral view, and the pronotal sides are entirely smooth, whereas in *S. piriformis*, these structures present different proportions and sculpturing patterns. These differences support the recognition of *S. brasiliensis* as a distinct species within the genus. The results contribute to the taxonomic knowledge of Eucoilinae and expand the geographic distribution of *Steleucoela*, reinforcing the importance of faunistic surveys in revealing new species in Neotropical regions (Gallardo *et al.*, 2003a).

12. *Muscidifurax Raptorellus* Kogan & Legner, 1970 (Hymenoptera: Pteromalidae) in Brazil

Chicken manure samples yielded dipteran pupae and associated parasitoids, enabling the identification of *M. raptorellus* as a parasitoid species present in Brazil. This represents the first record of this species in the country. A total of 80 fecal samples were collected from poultry facilities and processed in the laboratory. The pupae were obtained using the flotation method and individually maintained in gelatin capsules until the emergence of flies or parasitoids.

The specimens of *M. raptorellus* were identified after emergence and confirmed by specialists. Ten (10) pupae of *F. pusio* were collected, from which two (2) specimens of *M. raptorellus* emerged, representing 20.0% parasitism rate. This value indicates that the species is established in the environment, although at low frequency (Figure 25).



Figure 25: Parasitism rate of *Muscidifurax raptorellus* in dipteran pupae. Illustration showing parasitoid development and emergence from the host pupa. Representation of gregarious behavior with multiple individuals developing in a single host

Muscidifurax raptorellus is known to occur in two biological forms, one gregarious and one solitary, depending on geographic distribution. In Brazil, the occurrence observed contributes to the knowledge of its distribution and potential use in biological control programs targeting synanthropic flies associated with poultry production systems.

In Brazil, *F. pusio* was found parasitized by the following parasitoids: *Eurytoma* sp., *P. vindemmiae*, *P. egeria*, *S. drosophilae*, *S. nigra*, and *Spalangia* sp., indicating a diverse parasitoid community associated with dipteran hosts. The presence of *M. raptorellus* in poultry manure reinforces its importance as a natural enemy of filth flies and highlights its potential applicability in biological control programs in Brazil (Marchiori *et al.*, 2009).

13. Natural Enemies of *Chrysomya Albiceps* Collected in the States of Goiás and Minas Gerais, Brazil

A total of 5,314 pupae of *C. albiceps* were collected. Of these, 132 pupae were parasitized, yielding 820 parasitoid individuals and resulting in an overall parasitism rate of 2.5%. Parasitism rate occurred in different substrates, including human feces, carcasses, cattle kidney and liver, chicken viscera, fish, and cattle liver, demonstrating the ecological plasticity of parasitoid species associated with *C. albiceps*. The largest number of pupae was obtained from carcasses (3,941), followed by cattle kidney and liver (1,088), feces (168), fish (45), cattle liver (43), and chicken viscera (29) (Table 10).

Table 10: Parasitoids of *Chrysomya albiceps* collected in Goiás and Minas Gerais, Brazil. Distribution of substrates, parasitoid species, number of individuals, and parasitized pupae. Parasitism rate calculated from the number of parasitized pupae divided by the total pupae

Substrate	Total pupae	Parasitoid species	Years	No. individuals	Parasitized pupae	Parasitism rate (%)
Cattle liver	43	<i>Tachinobia</i> sp.	2013	30	1	2.3
Carcass	3941	<i>Paraganaspis egeria</i>	1999	5	5	0.1
Chicken viscera	29	<i>Brachymeria podagrica</i>	2001	4	4	13.8
Feces	168	<i>Hemencyrtus herbertii</i>	2003–2004	27	4	2.4
		<i>Trichopria</i> sp.	2004–2005	25	25	14.9
Fish	45	<i>Pachycrepoideus vindemmiae</i>	2006	30	30	66.7
		<i>Spalangia endius</i>	1999	7	7	15.6
Kidney liver	1088	<i>Nasonia vitripennis</i>	2005	692	56	5.1
Total	5314			820	132	2.5

Several parasitoid species were recorded, including *B. podagrica*, *H. herbertii*, *N. vitripennis*, *P. vindemmiae*, *P. egeria*, *S. endius*, *Tachinobia* sp., and *Trichopria* sp. In terms of parasitized pupae, *N. vitripennis* was the most frequent species, representing 42.4% (56/132) of the total parasitized pupae. In terms of emerged parasitoid individuals, *N. vitripennis* represented 84.4% (692/820), reflecting its gregarious behavior.

Parasitism rate varied among substrates and parasitoid species, with *P. vindemmiae* showing the

highest parasitism rate (66.7%) in fish, indicating greater efficiency or adaptation under the sampled conditions. Other species, such as *Trichopria* sp. and *B. podagrica*, also showed relevant parasitism rate levels depending on substrate and environmental conditions. The chi-square analysis showed a significant difference in parasitism rate among substrates ($\chi^2 = 1470.21$; $df = 5$; $p < 0.0001$), indicating that substrate type influenced parasitoid occurrence and success. This result should be interpreted with caution for substrates with small sample sizes, but it supports the observed variation in parasitism rate among substrates (Figure 26).

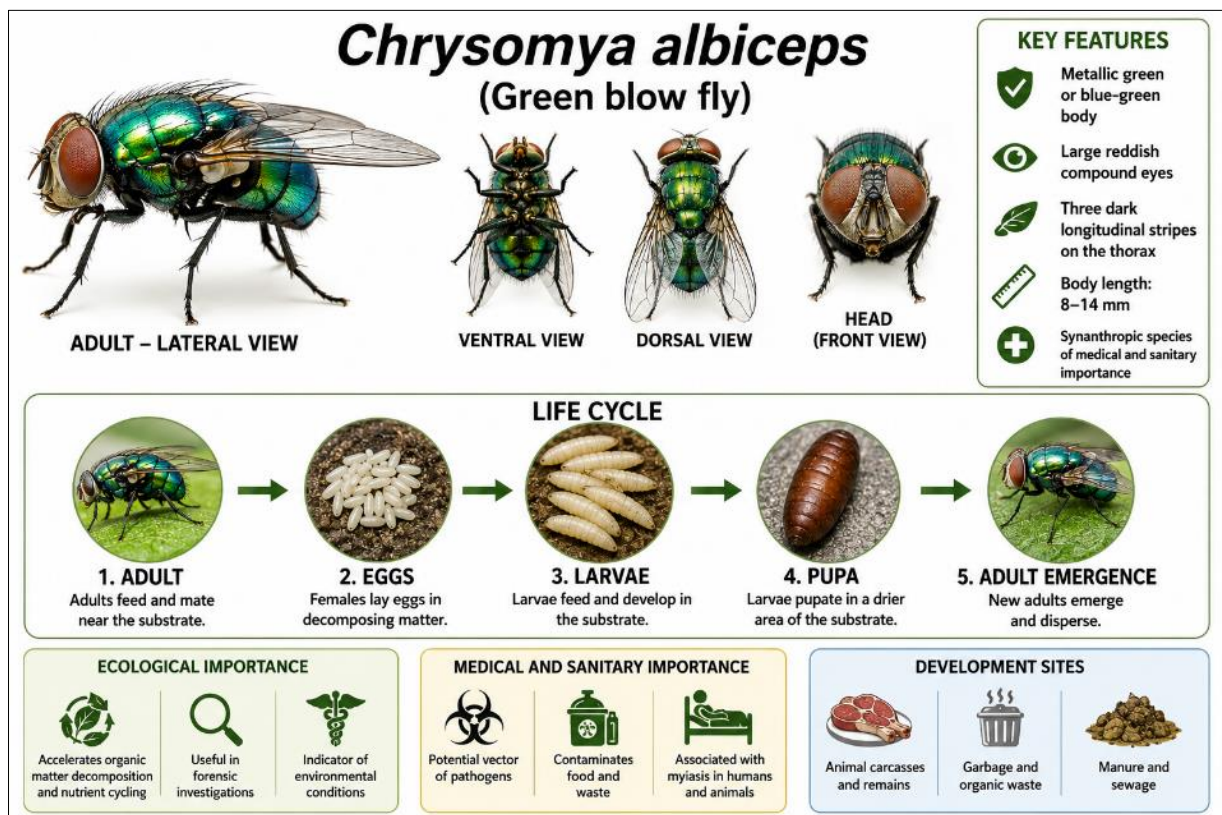


Figure 26: *Chrysomya albiceps* (green blow fly): adult morphology, life cycle, ecological importance, and medical-sanitary relevance. The figure highlights diagnostic features of the adult, developmental stages in decomposing substrates, and the role of this synanthropic species in organic matter decomposition, forensic investigations, pathogen transmission, food contamination, and myiasis

The most frequent parasitoid in terms of emerged individuals, *N. vitripennis*, exhibited gregarious behavior, which explains the high number of adults obtained from a smaller number of parasitized pupae. In contrast, *P. vindemmiae* is considered a solitary parasitoid and showed the highest parasitism rate among the recorded associations. These results demonstrate that parasitoid species associated with *C. albiceps* vary according to substrate and environmental conditions, highlighting their ecological importance and potential use in biological control programs targeting synanthropic flies (Marchiori, 2015a).

14. Arthropods (Diptera and Hymenoptera) Associated with Pig Carcasses in Itumbiara, Southern Goiás

The total parasitism rate reported for the carcass-associated pupae was 0.40%. In *C. albiceps* pupae, the parasitism rate was 0.30%, whereas in *Ophyra* sp. pupae it was 1.98%. *Pachycrepoideus vindemmiae* and *P. egeria* each showed a low parasitism rate, approximately 0.11%, and *S. endius* showed 0.16%. A total of 4,421 Diptera and Hymenoptera individuals were recorded in the carcass experiment, with 1,361 individuals (30.8%) obtained from the pasture carcass and 3,060 individuals (69.2%) from the forest carcass. A greater number of individuals and species were obtained in the forest, probably due to higher humidity during December, which may have favored carcass decomposition and arthropod development (Table 11).

Table 11: Abundance and relative frequency of Diptera and Hymenoptera collected from pig carcasses exposed in pasture and forest at the Faculty of Agronomy Farm in Itumbiara, Goiás, Brazil

Taxonomic group	Carcass Pasture	Frequency (%)	Carcass Forest	Frequency (%)
Diptera				
<i>Chrysomya albiceps</i>	1331	97.7	2610	85.7
<i>Fannia pusio</i>	30	2.3	62	2.0
<i>Hermetia illucens</i>	0	0	32	1.0
<i>Megaselia scalaris</i>	0	0	42	1.4
<i>Ophyra</i> sp.	0	0	302	9.9
Total	1361	100	3042	100
Hymenoptera				
<i>Pachycrepoideus vindemmiae</i>	0	0	6	33.4
<i>Paraganaspis egeria</i>	0	0	5	27.8
<i>Spalangia endius</i>	0	0	7	38.8
Total	0	0	18	100

Regarding abiotic factors related to carcass decomposition, relative humidity appeared to have a strong influence on the decomposition process. Based on Table 12, carcass decomposition took longer in October, when precipitation was lower, requiring 25 days, whereas in December, under higher precipitation, decomposition lasted only 9 days. In contrast, temperature did not appear to be a determining factor in

carcass degradation, because similar average temperatures were recorded in both months. The chi-square analysis showed a significant difference in total arthropod abundance between the pasture and forest carcasses ($\chi^2 = 652.93$; $df = 1$; $p < 0.0001$), with higher abundance in the forest. Dipteran species distribution also differed significantly between environments ($\chi^2 = 183.60$; $df = 4$; $p < 0.0001$).

Table 12: Climatic data recorded at the Faculty of Agronomy Farm in Itumbiara, Goiás, Brazil

Month	Average temperature (°C)	Precipitation (mm)	Local (environment)
October	26.4	61.6	Pasture
December	26.3	267.3	Forest

The higher abundance and richness observed in the forest area may be related to increased humidity and precipitation, which can accelerate carcass decomposition and favor the development of immature Diptera and their associated parasitoids. The presence of Hymenoptera only in the forest carcass reinforces the importance of environmental conditions for parasitoid occurrence.

Knowledge of the insect fauna associated with animal carcass decomposition, together with climatic

conditions, is important because these data can be applied in forensic investigations to support the interpretation of decomposition processes and the estimation of the postmortem interval (Marchiori *et al.*, 2000).

15. First Report of *Tachinobia Repanda* Bouček, 1977 (Hymenoptera: Eulophidae) In Brazil

This study reports the first occurrence of *T. repanda* in Brazil, contributing to the knowledge of the geographical distribution of this parasitoid. Eleven pupae

of *Oxysarcodexia* sp. were obtained, and 10 adult individuals of *T. repanda* emerged from one parasitized pupa, demonstrating gregarious parasitic behavior. The observed parasitism rate was 9.1%.

The results indicate that *T. repanda* acts as a pupal endoparasitoid, with multiple individuals developing within a single host, a characteristic of gregarious parasitoids. This reproductive strategy may increase the parasitism rate efficiency and enhance its potential as a biological control agent. The host,

Oxysarcodexia sp., is commonly associated with decomposing organic matter and carcasses, environments that favor the occurrence of Diptera and their parasitoids.

Tachinobia repanda was described by Bouček from Papua New Guinea. Its known distribution includes Southeast Asia, including Papua New Guinea, Indonesia, Malaysia, India, the Solomon Islands, and Thailand. The species has also been recorded in the southern United States and Cuba, where it is likely invasive (Figure 27).

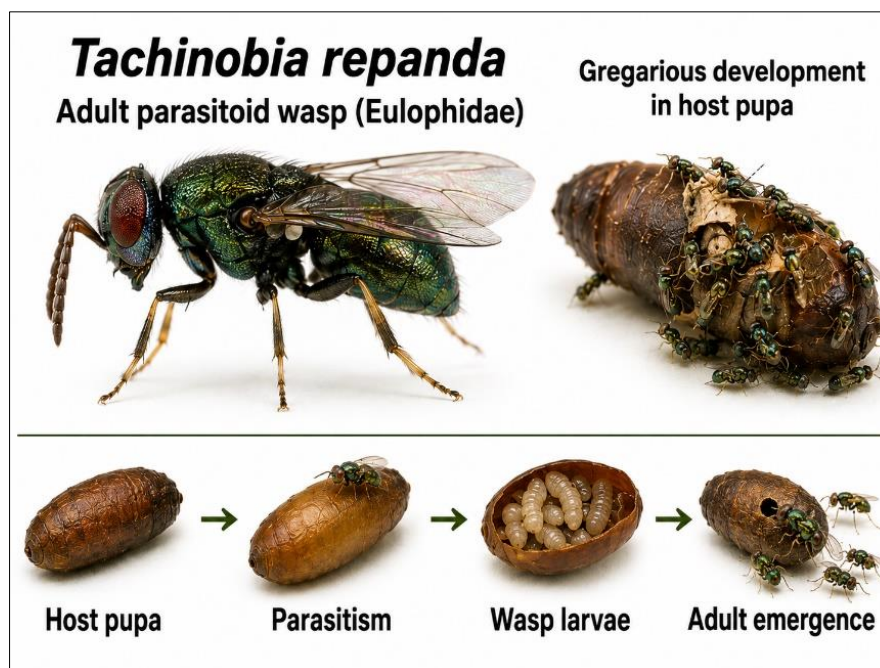


Figure 27: Adult parasitoid wasp of the family Eulophidae associated with dipteran hosts. The species *Tachinobia repanda* shows gregarious development, with multiple parasitoid individuals emerging from a single host pupa

The methodology used, including baited traps and subsequent recovery of pupae from sand, proved effective for obtaining parasitoids. The individualization of pupae in gelatin capsules allowed accurate observation of emergence and host-parasitoid associations. The presence of *T. repanda* in Brazil expands its known geographical distribution and reinforces the importance of faunistic surveys in different environments to improve knowledge of parasitoid diversity. From an applied perspective, parasitoids such as *T. repanda* may play a relevant role in the natural regulation of dipteran populations. Because some dipteran species are of medical and sanitary importance, identifying their natural enemies is essential for the development of biological control strategies (Marchiori, 2021a).

16. *Tachinobia* sp. (Hymenoptera: Eulophidae) as Parasitoid of *Sarcodexia Lambens*

A total of 6 pupae were obtained from *S. lambens*, of which 1 pupa produced 12 specimens of *Tachinobia* sp. The parasitism rate was 16.7%. The success of parasitism rate can be influenced by resource availability, host density, and the searching capacity of parasitoids. The Sarcophagidae are distributed worldwide and comprise about 2,600 known species. The Neotropical fauna of Sarcophagidae is highly diverse, with over 750 described species; despite this diversity, little is known about the biology of the group in this region. *Peckia* is considered a senior synonym of *Sarcodexia* Townsend, 1892 (Diptera: Sarcophagidae), with the latter maintained as a valid subgenus, resulting in the new generic combination *S. lambens* (Figure 28).

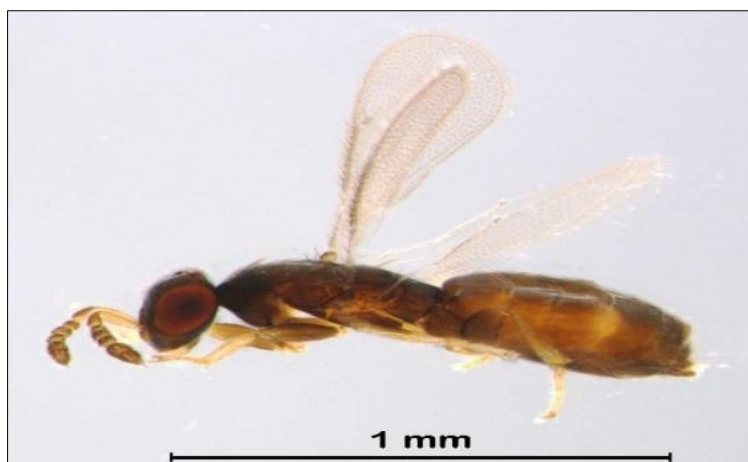


Figure 28: Adult *Tachinobia* sp. Lateral view showing characteristic coloration and body morphology. They are gregarious parasitoids, meaning that several wasps emerge from a single host pupa

This species, which ranges from the southern United States to Argentina, is a nonspecific parasite and has been recorded parasitizing birds, mollusks, scorpions, and insects, and has also been documented as a myiasis-causing fly in some vertebrates, including humans. This species has also been reported as a host of other parasitic insects.

Among the methods for controlling flies, chemical insecticides are the most widely used. However, their effectiveness may decrease as populations gradually develop resistance. This growing resistance highlights the need to introduce alternative control strategies, such as biological control. It is

possible to control these insects using such approaches (Marchiori, 2021b).

17. First Report of *Paraganaspis Egeria* Parasiting Horn Fly, *Haematobia Irritans* in Brazil

The Figitidae (Hymenoptera) are parasitoids of Muscomorpha (Diptera) immatures. The Eucoilidae are the most common Cynipoidea in the Neotropical region. It is supposed that the genus *Paraganaspis* Diaz, Gallardo & Walsh, 1996, is extensively distributed in the Neotropical region. *Paraganaspis egeria* parasiting *S. occidua* has been recorded in many localities in Argentina and in the Brazilian states of São Paulo and Mato Grosso do Sul (Figure 29).



Figure 29: Pupa of *Haematobia irritans* parasitized by the parasitoid *Paraganaspis egeria* in Brazil. These are small to minute parasitoid wasps that act as endoparasitoids of Cyclorrhapha Diptera larvae

The horn fly, *H. irritans*, was first found in Northern Brazil in the early 1980 and now infests cattle throughout South America. It is an important bovine pest in many countries, including Brazil. During the study period, 8.3% of the parasitism rate in the horn fly was caused by *Spalangia* spp., Diapriidae, and Eucoilinae.

Two (2) from 718 (0.26%) horn fly pupae collected in the middle of April 1994, at the beginning of the cold and dry season (19.8 °C and rainfall of 62.5mm³), were parasitized by an Eucoilinae, now confirmed as belonging to the species *P. egeria* (Figure 30) (Peretti, 2026).

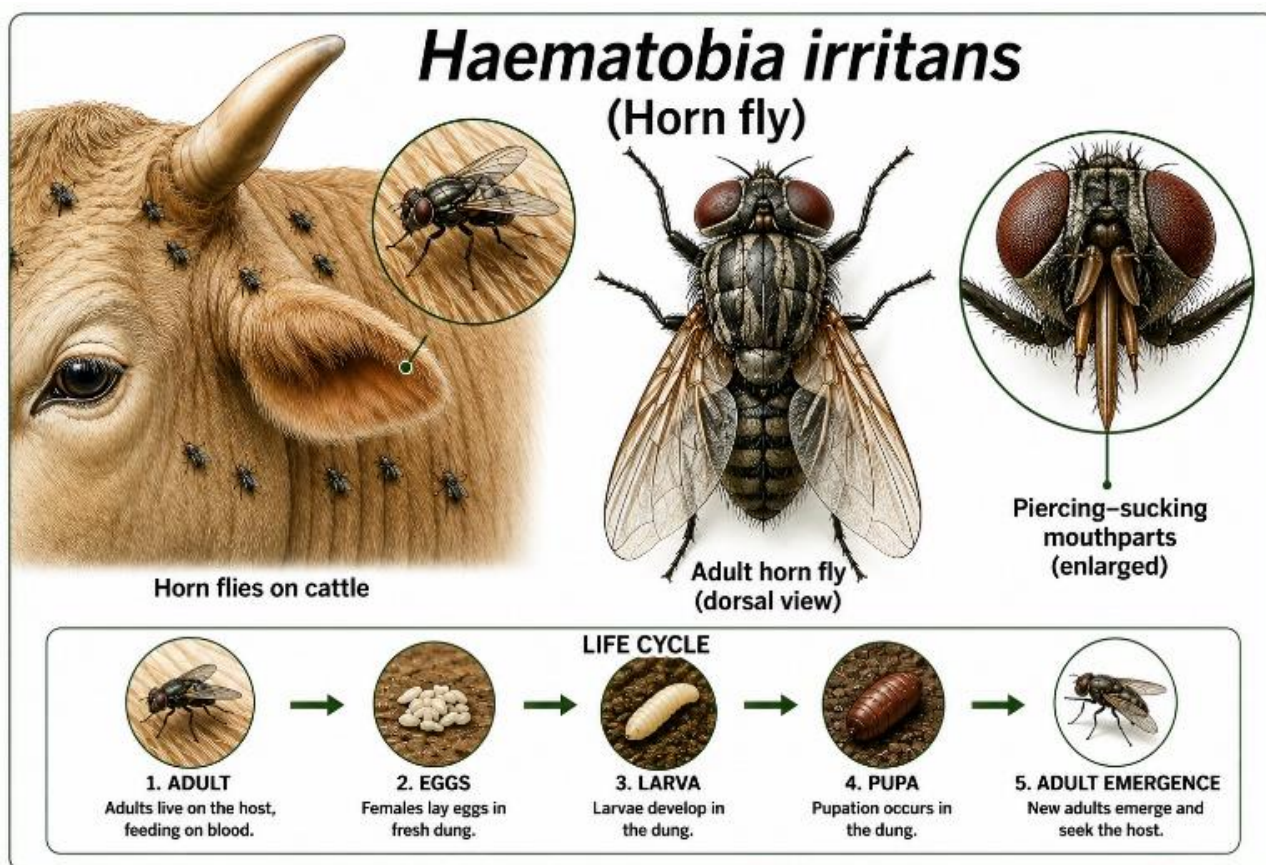


Figure 30: *Haematobia irritans* (horn fly) associated with cattle. The figure illustrates adult horn flies on the bovine host, a detailed view of the adult body morphology, and an enlargement of the piercing-sucking mouthparts used for blood feeding. This hematophagous ectoparasite is of veterinary and economic importance because it causes irritation, stress, reduced weight gain, and lower milk production in cat

This is the first record of *P. egeria* parasitizing the horn fly in the Neotropical region. Despite the low parasitism rate verified by *P. egeria* in the horn fly, this relationship merits further investigation (Marchiori *et al.*, 2001a).

18. *Trichopria* sp. in *Haematobia Irritans* Pupae in Brazil

A total of 350 pupae emerged, from which 13 specimens of the parasitoid *Trichopria* sp. emerged. The parasitism rate obtained was 3.7%, probably due to variations in the quality and availability of food resources or the densities of the hosts. This frequency of parasitism rate may also be showing greater specificity or affinity of the parasitoid *Trichopria* sp. for *S. occidua* pupae.

Trichopria sp. was recorded parasitizing pupae of *H. irritans* in Brazil. Adult parasitoids emerged from

pupae obtained from cattle dung collected in pasture areas, confirming the association of this diapiiid species with muscoid flies of veterinary importance under natural conditions. The presence of *Trichopria* sp. in this substrate suggests that this parasitoid is well adapted to environments rich in decomposing organic matter, such as bovine feces, where *H. irritans* completes its immature development.

Diapriidae species are recognized as endoparasitoids of Cyclorrhapha Diptera, attacking larval stages and completing their development within the host puparium. The occurrence of this parasitoid indicates its potential contribution to the natural biological control of *H. irritans*, an economically important ectoparasite of cattle. The interaction between parasitoids and dipteran hosts in dung pats plays a relevant ecological role, helping regulate fly populations in livestock systems (Figure 31).



Figure 31: *Trichopria* sp., wasps are characterized by a median facial protuberance from which the antennae arise, a feature visible in this specimen. The small body size and reduced wing venation suggest placement within the subfamily Diapriinae. Source: Photo#15884, Richard Leung, Iowa State University, and <https://www.bugguide.net/node/view/15884>

Although quantitative data on parasitism rate levels were not detailed in this study, the record of *Trichopria* sp. expands the knowledge of parasitoid diversity associated with *H. irritans* in Brazil. These findings reinforce the importance of surveys of natural enemies in pasture ecosystems and highlight the potential use of native parasitoids in integrated pest management programs (Marchiori, 2001b).

19. *Aphaereta* sp. as a Natural Enemy to *Peckia Chrysostoma* in Brazil

During the period from March to September in 2001, 374 specimens of *Aphaereta* sp. were collected from 26 pupae of *P. chrysostoma*. From the first to twelfth pupae were found 8, 10, 22, 27, 28, 29, 31, 31, 40, 42, 47, and 60 specimens, respectively. The parasitism rate (12/ 26) 79.5%. The results demonstrate that *Aphaereta* sp. is an endoparasitoid associated with synanthropic Diptera developing in decomposing organic substrates. Its gregarious habit suggests an efficient exploitation of host resources, allowing the development of several individuals within a single puparium. This reproductive strategy may enhance its potential as a biological control agent.

Aphaereta sp. is an important component in cattle excrement, attacking pupae of *H. irritans* in the Mississippi estate. In fact, the *Aphaereta* sp. has attracted the attention of a large number of investigators as a potential biological control, because of its wide distribution and host range. This species was a dipterous parasitoid; it developed in bovine liver in typical wood areas in the state of Goiás.

The occurrence of *Aphaereta* sp. in pupae of *P. chrysostoma* represents the first record of this host-parasitoid association in Brazil. This finding expands the known host range of the genus and highlights its ecological importance in environments associated with animal production and organic waste. Furthermore, the presence of this parasitoid in cattle-associated environments reinforces its role in the natural regulation of dipteran populations of sanitary and veterinary importance. Species of Alysiniinae are known to parasitize Diptera, and their occurrence in such habitats suggests potential applications in integrated pest management programs (Figure 32) (Marchiori *et al.*, 2003b).

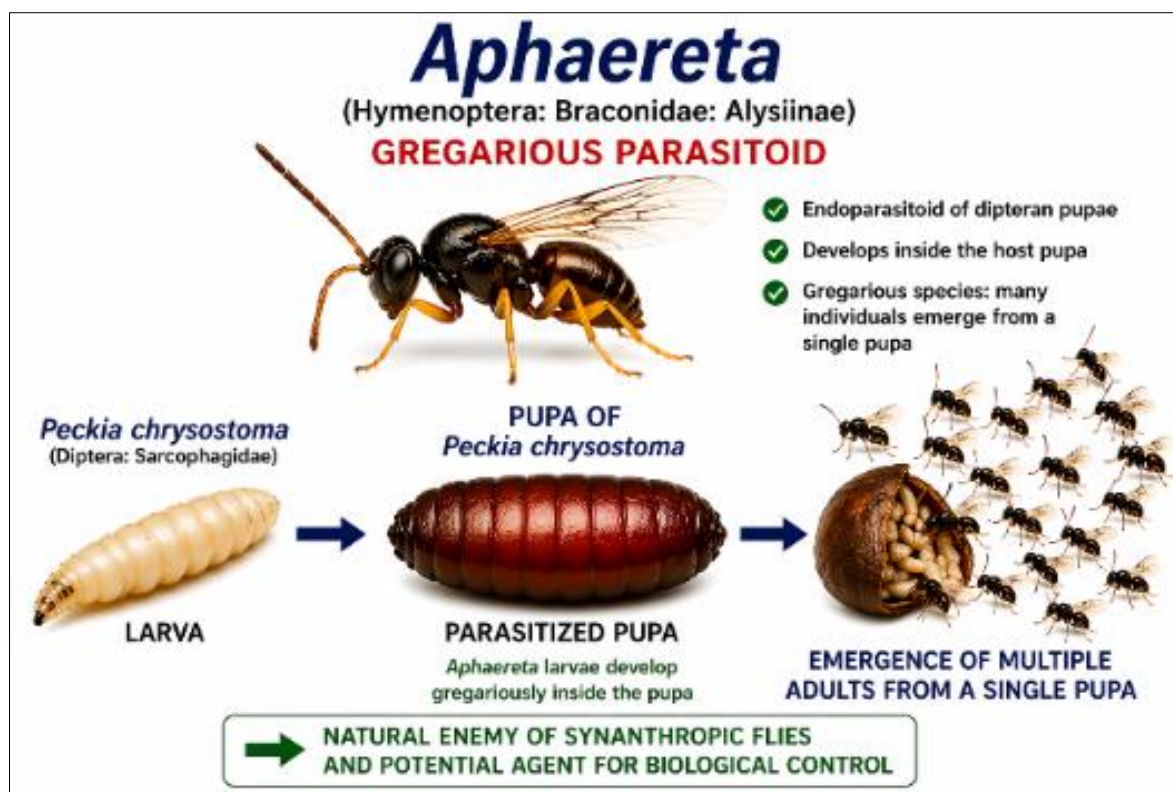


Figure 32: *Aphaereta* parasitizing pupae of *Peckia chrysostoma*. The species exhibits gregarious development, with multiple individuals emerging from a single host pupa. This behavior highlights its role as a natural enemy of synanthropic flies and its potential in biological control

20. First Record of *Leptopilina Boulardi* Parasitizing *Zaprionus Indianus* Gupta (Diptera: Drosophilidae) in Brazil

Three (3) specimens of *L. boulardi* were obtained from 139 pupae of *Z. indianus*, resulting in a parasitism rate of 2.2% (3/139). The parasitoids were collected using fruit-baited traps in Itumbiara, Goiás, Brazil (Figure 33).



Figure 33: *Leptopilina boulardi*, a parasitoid of Drosophilidae larvae. This species oviposits in larvae of *Zaprionus indianus*, where its development occurs. It plays an important role as a natural enemy in regulating fly populations. Source: *Leptopilina* Wikipedia and <https://alchetron.com/Leptopilina>

The low parasitism rate observed may be associated with factors such as host density, resource availability, and the parasitoid's searching efficiency. The limited number of individuals suggests that *L.*

boulardi may occur at low frequencies in this environment or that environmental conditions were not optimal for its development (Figure 34).

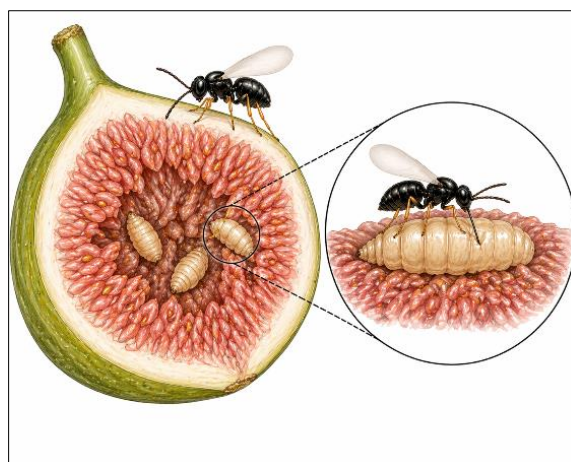


Figure 34: Larvae of *Zaprionus indianus*, hosts of the parasitoid *Leptopilina boulandi*. The larvae develop within fruit substrates, where the parasitism occurs. This interaction highlights the ecological relationship between Drosophilidae hosts and parasitoid wasps

This study represents the first record of *L. boulandi* parasitizing *Z. indianus* in Brazil, expanding the known host range and geographic distribution of this parasitoid species. The association between *L. boulandi*

and Drosophilidae hosts highlights its ecological importance as a natural enemy of fruit-infesting flies (Figures 35-36) (Kacsoh *et al.*, 2012; Fartyal *et al.*, 2014; Martínez *et al.*, 2017).

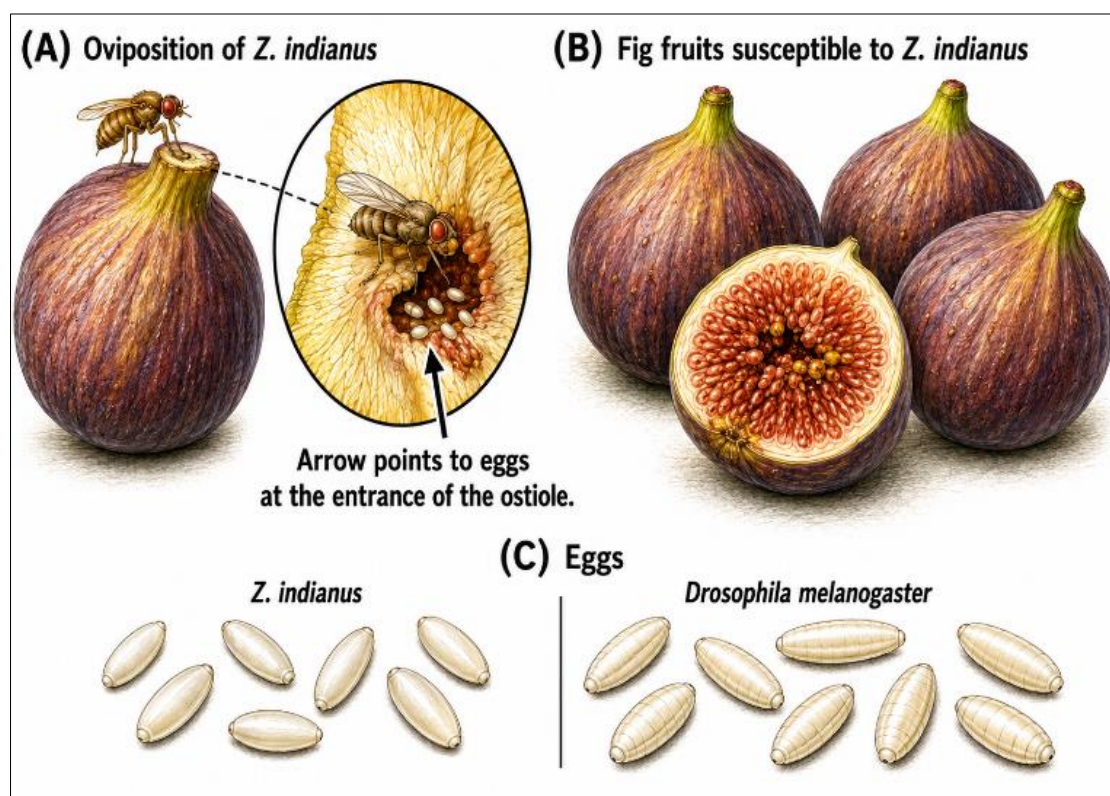


Figure 35: Oviposition and egg morphology of *Zaprionus indianus* in fig fruits. The figure illustrates oviposition at the fig ostiole, fig fruits susceptible to infestation, and comparative egg morphology of *Z. indianus* and *Drosophila melanogaster*. The arrow indicates eggs deposited at the entrance of the ostiole

The use of chemical control methods for dipteran populations may lead to resistance and environmental impacts, reinforcing the importance of alternative strategies such as biological control. In this

context, parasitoids like *L. boulandi* may contribute to the natural regulation of dipteran populations, particularly in agroecosystems associated with fruit production (Figure 36) (Marchiori *et al.*, 2003d).

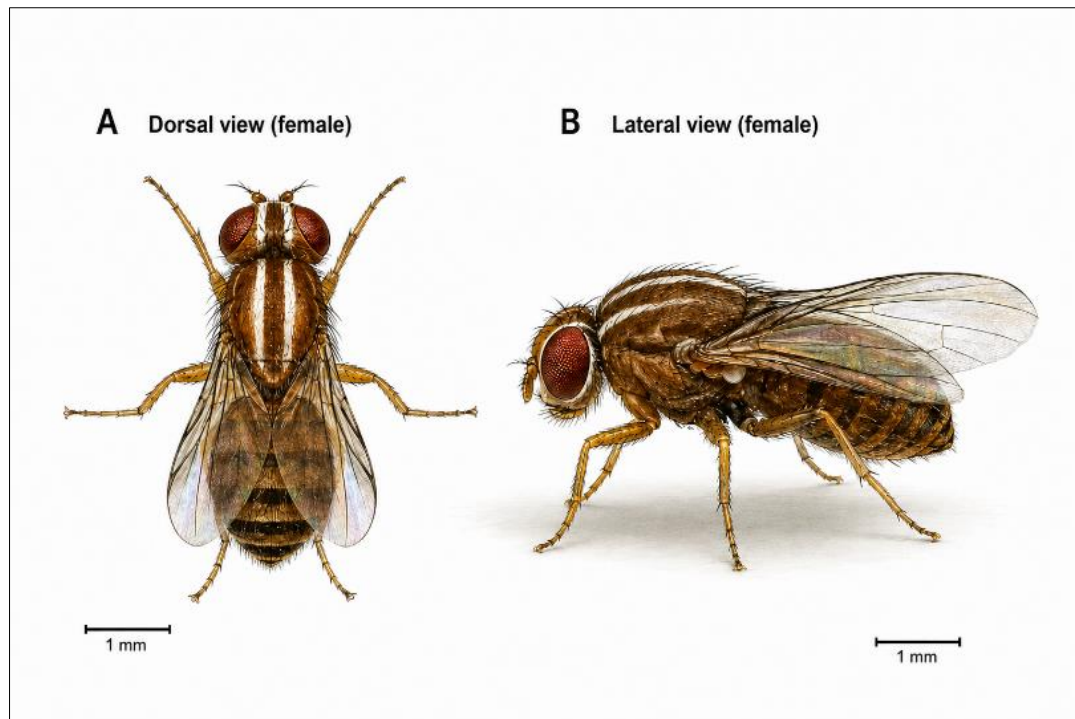


Figure 36: Adult specimens of *Zaprionus indianus* showing diagnostic morphological features. (A) Dorsal view of a female, highlighting the two conspicuous longitudinal white stripes on the head and thorax. (B) Lateral view of a female adult, showing the general body morphology, wing position, and reddish compound eye. Scale bars: 1 mm

21. Parasitoids of Diptera Collected in the Urban Area of Goiânia, Goiás, Brazil

A total of 358 dipteran pupae were collected in the urban area of Goiânia, Goiás, Brazil, from which 82 parasitoid individuals emerged. Sixty pupae were parasitized, resulting in an overall parasitism rate of

16.8%. This result may be related to the occurrence of gregarious parasitoids and to the availability of suitable hosts in urban environments. The parasitoids recorded were associated mainly with *S. lambens* and *S. nudiseta* (Table 13).

Table 13: Parasitoids collected in urban areas of Goiás, Brazil. The table presents host species, number of pupae, parasitoid taxa, number of individuals, number of parasitized pupae, and parasitism rate

Taxonomic group	Hosts species	Number of pupae	Parasitoids species	Frequency (%)	Number of individuals	Number of pupae parasitized	Parasitism rate (%)
Calliphoridae	<i>Chrysomya albiceps</i>	08	-	2.2	-	-	-
Fanniidae	<i>Fannia pusio</i>	155	-	43.3	-	-	-
Muscidae	<i>Synthesiomyia nudiseta</i>	33	<i>Pachycrepoides vindemmiae</i>	9.2	4	4	12.1
Sarcophagidae	<i>Peckia chrysostoma</i>	25	-	7.0	-	-	-
	<i>Sarcodexia lambens</i>	137	<i>Brachymeria podagrica</i>	38.3	53	53	38.7
		-	<i>Hemencyrtus herbertii</i>	-	13	1	0.7
		-	<i>Pachycrepoides vindemmiae</i>	-	1	1	0.7
		-	<i>Tachinobia</i> sp.	-	11	1	0.7
Total		358		100	82	60	16.8

Among the parasitoids collected, *B. podagrica* was the most abundant species, with 53 individuals emerging from 53 parasitized pupae of *S. lambens*. This species represented 64.6% of the parasitoid individuals

collected and 88.3% of the parasitized pupae recorded in the study. Other parasitoids were recorded in lower abundance: *Hemencyrtus herbertii*, with 13 individuals emerging from one parasitized pupa; *P. vindemmiae*,

with five individuals in total; and *Tachinobia*, with 11 individuals emerging from one parasitized pupa.

The chi-square analysis showed a highly significant difference between parasitized and non-parasitized pupae ($\chi^2 = 158.61$; $df = 1$; $p < 0.0001$), with non-parasitized pupae being more frequent. Parasitism rate also differed significantly among host species ($\chi^2 = 95.47$; $df = 4$; $p < 0.0001$), indicating that parasitism rate was concentrated mainly in *S. lambens* and *S. nudiseta*. In addition, parasitoid abundance differed significantly among species ($\chi^2 = 70.39$; $df = 3$; $p < 0.0001$), confirming the predominance of *B. podagrica* in the urban area.

The relatively low diversity of parasitoid species observed may be associated with urbanization, which can reduce the availability of ecological niches and limit species richness. However, the dominance of *B. podagrica* suggests that this species is well adapted to urban conditions, possibly due to host availability, resource quality, and its association with muscoid Diptera of medical and veterinary importance. The presence of parasitoids in urban environments highlights their ecological role as natural enemies capable of regulating dipteran populations. Considering that many synanthropic flies are vectors of pathogens, parasitoids may represent useful agents in biological control strategies in urban and agricultural systems (Figure 37) (Marchiori, 2015; van Noort, 2026).

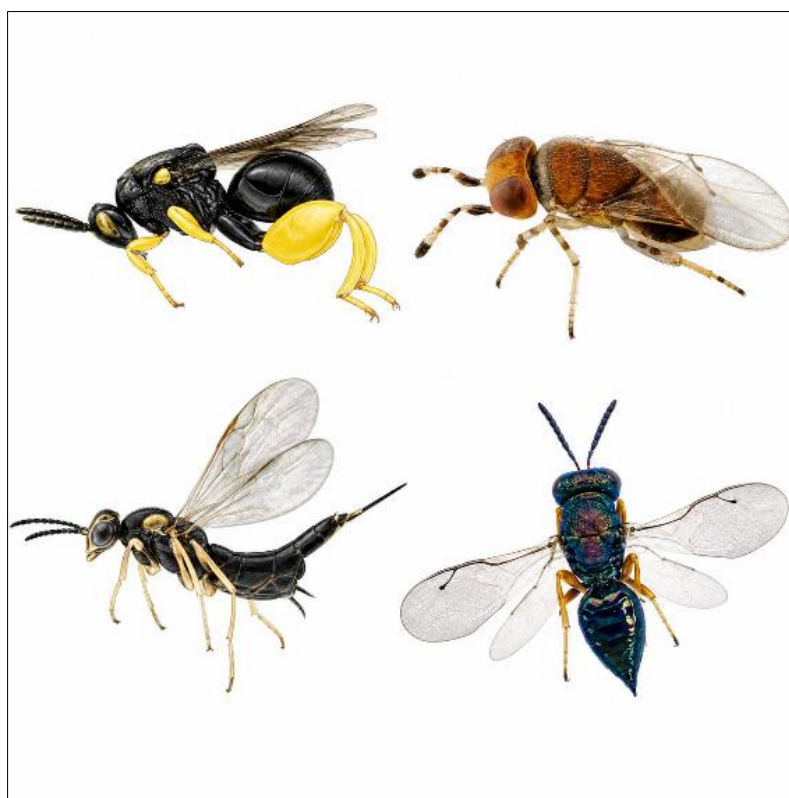


Figure 37: Representative parasitoid families associated with Diptera hosts in urban environments. Chalcididae, Encyrtidae, Eulophidae, and Pteromalidae are shown as natural enemies of synanthropic flies. Modified from source: Photo#16762/Richard Leung/Iowa State University and Simon van Noort (Iziko South African Museum)

22. Natural Occurrence of Dipteran Parasitoids Collected on Poultry Farms

A total of 2,815 synanthropic Diptera were collected from poultry manure, from which 795 parasitoids were obtained, resulting in an overall parasitism rate of 28.2%. These values demonstrate the significant occurrence of natural enemies associated with dipteran populations in poultry production systems. The parasitoid community comprised several taxa, and *Pachycrepoideus vindemmiae* was the most abundant

species, with 367 individuals, corresponding to 46.2% of the collected parasitoids. This higher frequency may be related to host availability, seasonal conditions, searching capacity, and the competitive ability of this parasitoid, although these factors were not experimentally tested in the original study. The diversity of parasitoids recorded highlights the complexity of parasitoid assemblages associated with synanthropic Diptera in poultry environments (Table 14).

Table 14: Parasitoids of synanthropic Diptera collected from poultry manure in Morrinhos, Goiás, Brazil. The table shows the monthly distribution of parasitoid species and the total number of individuals recorded from August to December

Parasitoids species	August	September	October	November	December	Total
Encyrtidae						
<i>Tachinaephagus zealandicus</i>	0	10	0	0	0	10
Pteromalidae:						
<i>Muscidifurax raptorellus</i>	2	101	0	0	0	103
<i>Nasonia vitripennis</i>	70	90	25	3	3	191
<i>Pachycrepoideus vindemmiae</i>	39	251	63	6	8	367
<i>Spalangia cameroni</i>	0	0	2	0	0	2
<i>Spalangia drosophilae</i>	0	0	2	0	0	2
<i>Spalangia endius</i>	14	40	10	0	3	67
<i>Spalangia nigra</i>	0	16	0	0	0	16
<i>Spalangia nigroaenea</i>	0	0	2	0	0	2
<i>Spalangia</i> sp.	12	10	3	0	0	25
Total	137	528	107	9	14	795

The highest number of parasitoids occurred in September, with 528 individuals, followed by August, with 137 individuals. Together, August and September accounted for 665 parasitoids, representing 83.6% of the total collected. Lower abundance was recorded from October to December, when precipitation and humidity increased. Excess moisture in poultry manure may have hindered host development and reduced parasitoid

emergence. The monthly distribution of parasitoids differed significantly among sampling months ($\chi^2 = 1150.15$; $df = 4$; $p < 0.0001$), indicating strong temporal variation in parasitoid occurrence. When the dry months were compared with the wetter months, the difference was also significant ($\chi^2 = 360.03$; $df = 1$; $p < 0.0001$), confirming that parasitoid abundance was higher during the dry period (Table 15).

Table 15: Precipitation, mean temperatures, and mean monthly humidities in the Morrinhos region, Goiás, Brazil. The values were obtained from the meteorological station of the State University of Goiás (INMET/UEG), Morrinhos, Goiás

Weather Data	August	September	October	November	December
Minimum temperature (°C)	13.1	16.0	19.1	19.7	19.2
Maximum temperature (°C)	30.9	33.0	33.6	31.1	29.4
Precipitation (mm)	0	0	22.2	100.0	285.8
Minimum humidity	26.0	26.0	34.4	47.7	59.5
Maximum humidity	46.0	40.2	51.1	70.0	74.3

The highest parasitism rates were observed with *P. vindemmiae*, likely due to its polyphagous habit and ability to exploit several dipteran hosts. This species showed the highest parasitism rate in *M. domestica*, with 347 parasitized pupae from 2,083 hosts, corresponding to 16.7%. *Pachycrepoideus vindemmiae* also parasitized *C. megacephala*, *F. pusio*, *Palaeosepsis* sp., and *O. obesa*, demonstrating broad host use. The species *N.*

vitripennis and *M. raptorellus* were also important components of the parasitoid community, especially in association with *M. domestica*. The host-associated records listed in Table 16 account for 785 parasitoids; the additional 10 unidentified Hymenoptera specimens recorded in Table 14 were not assigned to a specific host in Table 16.

Table 16: Parasitoids and their synanthropic Diptera hosts collected from poultry manure in Morrinhos, Goiás, Brazil. The table presents host species, number of individuals, parasitoid taxa, number of parasitized pupae, and of parasitism rate

Diptera	Number of hosts	Parasitoids species	Parasitized pupae	Parasitism rate (%)
<i>Chrysomya megacephala</i>	500	<i>Nasonia vitripennis</i>	3	0.6
		<i>Pachycrepoideus vindemmiae</i>	3	0.6
		<i>Spalangia endius</i>	3	0.6
<i>Fannia pusio</i>	42	<i>Muscidifurax raptorellus</i>	2	3.8
		<i>Pachycrepoideus vindemmiae</i>	2	3.8
<i>Musca domestica</i>	2083	<i>Muscidifurax raptorellus</i>	101	4.8
		<i>Nasonia vitripennis</i>	188	9.0
		<i>Pachycrepoideus vindemmiae</i>	347	16.7

Diptera	Number of hosts	Parasitoids species	Parasitized pupae	Parasitism rate (%)
		<i>Spalangia endius</i>	67	3.2
		<i>Spalangia nigra</i>	16	0.8
		<i>Spalangia nigroaenea</i>	2	0.1
		<i>Spalangia</i> sp.	15	0.7
		<i>Tachinaephagus zealandicus</i>	10	0.5
<i>Palaeosepsis</i> sp.	81	<i>Nasonia vitripennis</i>	2	2.5
		<i>Pachycrepoideus vindemmiae</i>	8	10.0
		<i>Spalangia drosophilae</i>	2	2.5
		<i>Spalangia</i> sp.	10	12.3
<i>Ornidia obesa</i>	93	<i>Pachycrepoideus vindemmiae</i>	2	2.2
		<i>Spalangia cameroni</i>	2	2.2
Other non-parasitized species	16	-	-	-
Total host-associated records	2815	-	785	27.9

Host-parasitoid associations varied among dipteran species. *Muscidifurax raptorellus* was associated with *F. pusio* and *M. domestica*; *N. vitripennis* was associated with *C. megacephala*, *M. domestica*, and *Palaeosepsis* sp.; *P. vindemmiae* was recorded in several hosts, including *C. megacephala*, *F. pusio*, *M. domestica*, *Palaeosepsis* sp., and *O. obesa*; *S. endius* was associated with *C. megacephala* and *M. domestica*; and *T.*

zealandicus was recorded in *M. domestica*. The chi-square analysis showed a significant difference in parasitism rate among host species ($\chi^2 = 273.26$; df = 5; $p < 0.0001$). The distribution of host-associated parasitoid taxa also differed significantly among hosts ($\chi^2 = 611.39$; df = 36; $p < 0.0001$), indicating that parasitoid occurrence was not randomly distributed among dipteran hosts (Figure 38).

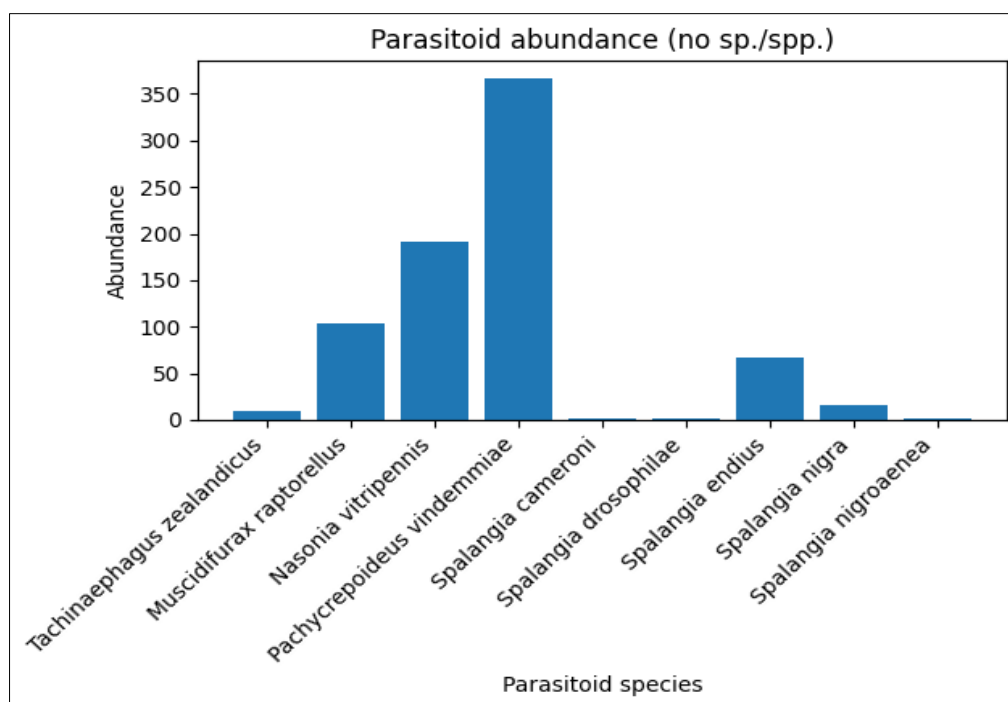


Figure 38: Abundance of parasitoid species associated with synanthropic Diptera collected from poultry manure in Morrinhos, Goiás, Brazil. *Pachycrepoideus vindemmiae* was the most abundant species, followed by *Nasonia vitripennis* and *Muscidifurax raptorellus*

Poultry farming in southern Goiás is an important economic activity, and the natural occurrence of parasitoids emerging from dipteran puparia in poultry manure is relevant for alternative fly management. These parasitoids may contribute to the natural regulation of synanthropic Diptera populations and support sustainable biological control strategies in poultry production systems (Marchiori and Miranda, 2011).

23. Natural Parasitism of *Musca Domestica* by Parasitoids in Organic Substrates in Itumbiara, Goiás, Brazil.

Pupae of synanthropic Diptera were collected from organic residues ("food waste") used for swine feeding in the municipality of Itumbiara, Goiás, Brazil. The material was transported to the laboratory, where pupae were obtained using the flotation method. Each

pupa was individually placed in gelatin capsules and maintained under laboratory conditions until the emergence of adult flies or parasitoids. Emerged specimens were identified using appropriate taxonomic keys.

A total of 312 pupae of *M. domestica* were collected from organic residues. From these, 68 parasitoids emerged, resulting in an overall parasitism rate of 21.8%. The natural parasitism of *M. domestica* by parasitoids in organic substrates in Itumbiara, Goiás, Brazil. e parasitoid species recorded were: *Pachycrepoideus vindemmiae* (52 individuals) and *S. endius* (16 individuals). *Pachycrepoideus vindemmiae* was the most abundant species, representing approximately 76.5% of the total parasitoids collected.

The occurrence of parasitoids associated with *M. domestica* in organic residues demonstrates the importance of these natural enemies in environments rich in decomposing organic matter. The predominance of *P. vindemmiae* is likely related to its polyphagous habit and high ecological adaptability, allowing it to exploit a wide range of hosts and substrates. Its competitive advantage during immature stages may also contribute to its higher frequency. Although less abundant, *S. endius* was also recorded, indicating the coexistence of parasitoid species with different biological strategies. This diversity contributes to the natural regulation of synanthropic Diptera populations. These findings highlight the potential of parasitoids as biological control agents in environments where organic waste accumulates, such as rural and peri-urban areas (Figure 39) (Marchiori and Silva, 2002b).



Figure 39: Collection site and developmental stages of synanthropic Diptera in organic residues (“food waste”) used for swine feeding. The image shows the container with decomposing material, pupae, adult flies, and parasitoids associated with the substrate. These materials were used for pupal collection and subsequent evaluation of parasitism under laboratory conditions

24. *Trichopria* sp. (Hymenoptera: Diapriidae) Parasitoids of Diptera Muscoidea

Although the family Diapriidae comprises numerous species, little is known about the biology of many of these parasitoids. The family includes the subfamilies Ambositrinae, Ismarinae, Belytinae, and Diapriinae. Oviposition may occur in the larval or pupal stage of the host. Most diapriids are parasitoids of Diptera, especially species belonging to Calliphoridae, Muscidae, Sarcophagidae, and Tachinidae.

According to Table 17, *Trichopria* sp. parasitized Diptera pupae at both collection sites, but the parasitism rate varied among host groups. At site 1, the highest parasitism rate was recorded in Sphaeroceridae, with 9 parasitized pupae from 88 pupae, corresponding to 10.2%. At site 2, the highest parasitism rate occurred in *S. occidua*, with 36 parasitized pupae from 1,060 pupae, corresponding to 3.4%. The overall parasitism rate was 0.7% at site 1 and 1.3% at site 2.

Table 17: Parasitism of Diptera pupae by *Trichopria* sp. at two collection sites. The table presents host species, number of pupae, parasitized pupae, and parasitism rates, highlighting differences between sites and host preference

Parasitoid species	Frequency	Hosts species	N. of pupae	Parasitism rate (%)
Site 1 (Uberlândia, MG)				
<i>Trichopria</i> sp.	2	<i>Brontaea quadristigma</i>	720	0.3
	32	<i>Palaeosepsis</i> sp.	3759	0.9
	14	<i>Sarcophagula occidua</i>	4080	0.3
	9	Sphaeroceridae sp.	88	10.2
Total	57	-	8647	0.7
Site 2 (Itumbiara, GO)				
<i>Trichopria</i> sp.	2	<i>Brontaea quadristigma</i>	204	1.0
	40	<i>Palaeosepsis</i> sp.	4289	0.9
	36	<i>Sarcophagula occidua</i>	1060	3.4
	1	Sphaeroceridae	397	0.3
Total	79		5950	1.3

Complementary chi-square analyses showed significant differences in parasitism rate among host groups at both collection sites. At site 1, parasitism rate differed significantly among hosts ($\chi^2 = 132.97$; $df = 3$; $p < 0.0001$), mainly because Sphaeroceridae showed the highest parasitism rate. At site 2, parasitism rate also

differed significantly among hosts ($\chi^2 = 43.42$; $df = 3$; $p < 0.0001$), with the highest rate observed in *Sarcophagula occidua*. Overall parasitism rate also differed significantly between the two sites ($\chi^2 = 17.07$; $df = 1$; $p < 0.0001$), being higher in Itumbiara, Goiás, than in Uberlândia, Minas Gerais (Figure 40).

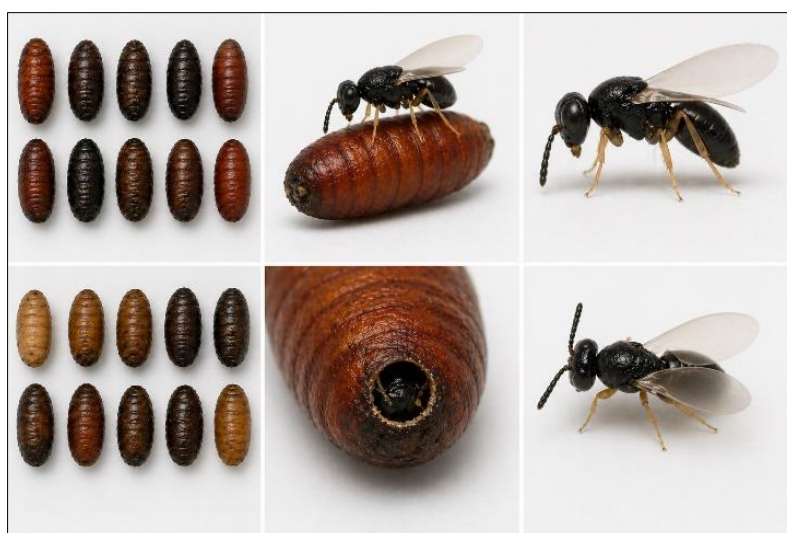


Figure 40: Parasitism of Diptera pupae by *Trichopria* sp. at two collection sites. The figure illustrates Diptera pupae collected from organic residues, female parasitoid activity on pupal hosts, emergence of adult parasitoids from puparia, and adult parasitoid morphology in lateral and dorsal views

Diapriidae are generally gregarious endoparasitoids of Diptera pupae; however, in the present study, *Trichopria* sp. behaved as a solitary parasitoid. The occurrence of *Trichopria* sp. in different host groups and at both collection sites indicates ecological plasticity and potential importance as a natural enemy of Diptera associated with decomposing organic substrates. In Brazil, species of *Trichopria* have been recorded in the states of Rio de Janeiro, São Paulo, Pará, and the Federal District. Therefore, this study extends the known geographic distribution of *Trichopria* sp. to Minas Gerais, in the Triângulo Mineiro region, and to southern Goiás (Marchiori *et al.*, 2000b).

25. Research on Diptera and Their Parasitoids (Insecta: Hymenoptera Parasitica) Collected in a Forest Area in Brazil

The data presented in Table 18 were checked against the text and showed internal consistency after correction of the total number of parasitized pupae. A total of 735 dipteran pupae were recorded from four substrates: bovine kidneys, chicken viscera, human feces, and fish. From these pupae, 89 were parasitized, resulting in an overall parasitism rate of 12.1% (Table 18).

Table 18: Diptera and their parasitoids collected from different substrates in a forest area in Brazil. The table presents host species, associated parasitoids, number of pupae, parasitized pupae, and parasitism rate

Substrate	Hosts species	Parasitoid species	Number of pupae	Parasitized pupae	Parasitism rate (%)
Bovine kidneys	<i>Fannia pusio</i>	<i>Pachycrepoideus vindemmiae</i>	44	2	4.5
Chicken viscera	<i>Chrysomya megacephala</i>	<i>Brachymeria podagrica</i>	14	2	14.3
	<i>Oxysarcodexia thornax</i>		67	10	15.0
	<i>Peckia chrysostoma</i>		174	34	19.5
Human feces	<i>Musca domestica</i>	<i>Pachycrepoideus vindemmiae</i>	274	18	6.6
	<i>Ophyra aenescens</i>	<i>Brachymeria podagrica</i>	46	11	24.0
	<i>Tricharaea occidua</i>	<i>Pachycrepoideus vindemmiae</i>	60	5	8.3
Fish	<i>Peckia chrysostoma</i>	<i>Brachymeria podagrica</i>	56	7	12.5
Total			735	89	12.1

The corrected total of parasitized pupae is 89, which agrees with the sum of the values shown in the table. Therefore, the overall parasitism rate was calculated as 89 parasitized pupae divided by 735 pupae, corresponding to 12.1%. This correction does not change the biological interpretation of the dataset, but improves the numerical consistency between the table and the text.

The distribution of parasitoids among substrates was coherent and biologically plausible. Chicken viscera showed the highest number of parasitized pupae, followed by human feces, fish, and bovine kidneys. At the host level, the highest parasitism rate was recorded for *O. aenescens* in human feces, followed by *P.*

chrysostoma and *O. thornax* in chicken viscera, and *C. megacephala* in chicken viscera.

The chi-square analysis showed a significant difference in parasitism rate among host species ($\chi^2 = 26.69$; $df = 7$; $p = 0.0004$), indicating that the parasitism rate was not evenly distributed among dipteran hosts. A second analysis also showed significant variation among substrates ($\chi^2 = 14.37$; $df = 3$; $p = 0.0024$), suggesting that substrate type influenced parasitoid occurrence. These results support the interpretation that host availability and substrate quality affected parasitoid activity in the forest environment (Figure 41).



Figure 41: Field collection of synanthropic Diptera and associated parasitoids in a forest environment. The figure shows traps installed in vegetation, decomposing substrates attracting flies, pupae maintained under laboratory conditions, and adult parasitoids emerging from pupae. These elements illustrate the host-parasitoid interaction under natural and controlled conditions

Variation in parasitism rate among substrates may be related to environmental and ecological factors, such as moisture, nutrient availability, odor intensity, and the degree of substrate decomposition. These factors can

directly affect the attraction and development of Diptera and, consequently, the availability of hosts for parasitoids. Substrates such as chicken viscera and human feces may provide favorable conditions for larval

development and pupal formation, increasing the opportunity for parasitism.

The presence of parasitoids in all substrates reinforces their ecological role as natural regulators of dipteran populations. This host-parasitoid interaction represents a dynamic system influenced by biotic and abiotic factors and contributes to the stability of insect communities associated with decomposing organic matter in forest environments (Marchiori, 2023).

26. *Hemencyrtus Herbertii* as Parasitoids of Dipteran Muscoid Flies Collected in Itumbiara, Goiás

Many genera of Encyrtidae exhibit polyembryonic reproduction, meaning that many

parasitoids emerge from a single host. Several species of this family have been successfully used in biological control programs. *Hemencyrtus herbertii* behaves as a larval parasitoid, developing internally within the host's body and emerging from the puparium.

A total of 511 Diptera pupae and 121 specimens of *H. herbertii* were collected from three different substrates. The total parasitism rate observed was 3.5% (18/511) (Table 19). Chicken was the substrate that attracted the largest number of *H. herbertii*, representing 58.0% of the individuals collected, and showed the highest parasitism rate, 2.3% (Table 19).

Table 19: Number of Diptera pupae collected from three different substrates, number of *Hemencyrtus herbertii* specimens emerged, and parasitism taxa

Bait	Hosts species	Number of individuals	Frequency of <i>Hemencyrtus herbertii</i>	Number of pupae	Parasitism rate (%)
Bovine liver	<i>Euboettcheria collusor</i>	31	3	1	3.2
	<i>Synthesiomia nudiseta</i>	55	20	2	3.6
Chicken	<i>Hemilucilia flavifacies</i>	16	14	3	18.8
	<i>Peckia chrysostoma</i>	280	61	9	3.2
Human feces	<i>Musca domestica</i>	20	5	1	5.0
	<i>Oxysarcodexia thornax</i>	109	18	2	1.8
Total	----	511	121	18	3.5

The high parasitism rate was probably influenced by variations in the quality and availability of resources, host density, and the gregarious behavior of the parasitoid. Regarding substrates, *H. herbertii* showed a preference for human feces ($\chi^2 = 2196.42$; $df = 24$; $p < 0.05$). Regarding hosts, *Hemencyrtus* sp. showed a preference for pupae of *O. thornax* (Marchiori *et. al.*, 2004c).

5.0. RESULTS OF THE REVIEWED ARTICLES

5.0.1 Chi-Square Analysis

The reviewed articles were checked by comparing the numerical information in the text with the corresponding tables. When the tables contained sufficient quantitative information, complementary chi-square analyses were added to support the interpretation of parasitism, host association, trap efficiency, substrate effects, and environmental differences. Tables were not reformatted, except for numerical corrections required to maintain consistency between the text and the original data.

Brachymeria podagrica collected in Brazil: A total of 2,430 dipteran pupae were recorded, with 395 pupae parasitized by *B. podagrica*, resulting in an overall parasitism rate of 16.3%. The highest parasitism rate occurred in *C. albiceps* (45.0%), followed by relevant parasitism rate in *P. chrysostoma* (18.5%) and *S. lambens* (18.4%). The chi-square test showed significant

variation in parasitism among host species ($\chi^2 = 55.74$; $df = 6$; $p < 0.0001$).

Hemencyrtus herbertii collected in Brazil: The dataset included 1,336 dipteran pupae and 31 pupae parasitized by *H. herbertii*, corresponding to an overall parasitism rate of 2.3%. The highest parasitism rate occurred in *S. lambens* (29.4%), followed by *H. flavifacies* (18.8%). The value for *E. collusor* was corrected to 0.2%. The chi-square test indicated significant differences in parasitism among host species ($\chi^2 = 88.29$; $df = 9$; $p < 0.0001$).

Parasitoids of *M. domestica* in rural and urban areas: In the rural area, 350 pupae of *M. domestica* were collected, with 24 parasitized pupae, resulting in 6.9% parasitism rate. In the urban area, 472 pupae were collected, with 42 parasitized pupae, resulting in 8.9% parasitism rate. Parasitism rate did not differ significantly between rural and urban areas ($\chi^2 = 1.13$; $df = 1$; $p = 0.287$). However, parasitoid species distribution differed significantly between environments ($\chi^2 = 14.96$; $df = 4$; $p = 0.0048$), with *P. vindemmiae* predominant in both areas.

Parasitoids of dipterans collected from animal excrement: In bovine feces, 628 pupae were recorded, and 77 were parasitized, resulting in 12.3% parasitism rate. In chicken feces, 3,116 pupae were recorded, and 310 were parasitized, resulting in 10.0% parasitism. The comparison between bovine and chicken feces was not significant ($\chi^2 = 3.02$; $df = 1$; $p = 0.082$). Host-parasitoid

associations were significant in bovine feces ($\chi^2 = 229.24$; $df = 66$; $p < 0.0001$) and chicken feces ($\chi^2 = 325.17$; $df = 40$; $p < 0.0001$).

Collection techniques for Diptera and Hymenoptera: This article was revised descriptively because it did not contain quantitative tables by trap type. Therefore, no chi-square analysis was applied. The corrected text emphasizes that Moericke traps, Malaise traps, metallic container traps, and pitfall traps differ in structure, selectivity, efficiency, and applicability, and that combined sampling methods are recommended for broader representation of Diptera and Hymenoptera communities.

Hymenoptera parasitoids collected using different trap types: A total of 400 parasitoids were recorded: 180 in metal container traps and 220 in pitfall traps. Pitfall traps collected more parasitoids overall, and the difference in abundance between trap types was significant ($\chi^2 = 4.00$; $df = 1$; $p = 0.0455$). Species distribution differed strongly between trap types ($\chi^2 = 322.04$; $df = 9$; $p < 0.0001$). *G. quadridentata* predominated in pitfall traps, whereas *P. vindemmiae* predominated in metal container traps.

Hemencyrtus herbertii associated with muscoid dipterans in Itumbiara, Goiás: The table recorded 511 pupae, 121 individuals of *H. herbertii*, and 18 parasitized pupae. Because this parasitoid is gregarious, the number of emerged parasitoids was higher than the number of parasitized pupae. The corrected overall parasitism rate was 3.5%. The chi-square analysis indicated significant variation in parasitism rate among host species ($\chi^2 = 12.05$; $df = 5$; $p = 0.0341$).

Natural enemies of *C. albiceps*: A total of 5,314 pupae of *C. albiceps* were recorded, with 132 parasitized pupae and 820 parasitoid individuals emerging. The corrected overall parasitism rate was 2.5%. Corrections were made for *H. herbertii* in feces (2.4%), *P. vindemmiae* in fish (66.7%), and *S. endius* in fish (15.6%). Parasitism rate differed significantly among substrates ($\chi^2 = 1470.21$; $df = 5$; $p < 0.0001$).

Arthropods associated with pig carcasses in pasture and forest: The corrected data showed 1,361 individuals from the pasture carcass and 3,060 from the forest carcass, corresponding to 30.8% and 69.2%, respectively. Hymenoptera were recorded only in the forest carcass. Total abundance differed significantly between environments ($\chi^2 = 652.93$; $df = 1$; $p < 0.0001$), and Diptera species distribution also differed significantly between pasture and forest ($\chi^2 = 183.60$; $df = 4$; $p < 0.0001$).

Parasitoids of Diptera in the urban area of Goiânia: The table recorded 358 pupae, 82 parasitoid individuals, and 60 parasitized pupae, corresponding to an overall parasitism rate of 16.8%. The parasitism rate

of *B. podagrica* in *S. lambens* was corrected to 38.7%. There was a significant difference between parasitized and non-parasitized pupae ($\chi^2 = 158.61$; $df = 1$; $p < 0.0001$), among host species ($\chi^2 = 95.47$; $df = 4$; $p < 0.0001$), and among parasitoid species ($\chi^2 = 70.39$; $df = 3$; $p < 0.0001$).

Natural occurrence of dipteran parasitoids on poultry farms: A total of 2,815 synanthropic Diptera were recorded, with 795 parasitized pupae, resulting in a corrected parasitism rate of 28.2%. *Pachycrepoideus vindemmiae* was the most abundant parasitoid. Monthly parasitoid distribution was significant ($\chi^2 = 1150.15$; $df = 4$; $p < 0.0001$), as was abundance among parasitoid species ($\chi^2 = 1797.42$; $df = 10$; $p < 0.0001$), parasitism rate among hosts ($\chi^2 = 273.26$; $df = 5$; $p < 0.0001$), and host-parasitoid association ($\chi^2 = 611.39$; $df = 36$; $p < 0.0001$).

Trichopria sp. parasitoids of Diptera Muscoidea: At site 1, 57 specimens of *Trichopria* were recorded from 8,647 pupae, resulting in 0.7% parasitism rate. At site 2, 79 specimens were recorded from 5,950 pupae, resulting in 1.3% parasitism rate. Corrected rates included 0.3% for *B. quadristigma* at site 1, 0.9% for *Palaeosepsis* sp., or *Palaeosepsis* spp., at site 2, and 3.4% for *S. occidua* at site 2. Host preference was significant at site 1 ($\chi^2 = 132.97$; $df = 3$; $p < 0.0001$), at site 2 ($\chi^2 = 43.42$; $df = 3$; $p < 0.0001$), and between sites overall ($\chi^2 = 17.07$; $df = 1$; $p < 0.0001$).

Diptera and parasitoids collected in a forest area: The table recorded 735 Diptera pupae and 89 parasitized pupae, resulting in a corrected overall parasitism rate of 12.1%. The original total of 90 parasitized pupae was corrected to 89. Parasitism differed significantly among host species ($\chi^2 = 26.69$; $df = 7$; $p = 0.0004$) and among substrates ($\chi^2 = 14.37$; $df = 3$; $p = 0.0024$), indicating that both host identity and substrate type influenced parasitoid occurrence.

Hemencyrtus herbertii as parasitoid of muscoid flies: The dataset included 511 Diptera pupae, 121 individuals of *H. herbertii*, and 18 parasitized pupae, confirming an overall parasitism rate of 3.5%. Chicken attracted the highest number of *H. herbertii* individuals (75; 62.0%), while bovine liver and human feces each recorded 23 individuals. Parasitism rate differed significantly among host species ($\chi^2 = 12.05$; $df = 5$; $p = 0.0341$), but the difference among substrates was not significant ($\chi^2 = 0.79$; $df = 2$; $p = 0.6736$).

The corrections confirmed that most datasets were suitable for complementary chi-square analyses, especially those involving parasitized and non-parasitized pupae, host species, substrates, environments, trap types, or monthly distributions. The main numerical corrections involved total parasitism rates, rounding of frequencies, distinction between the number of parasitoid individuals and the number of

parasitized pupae, and interpretation of gregarious parasitoids. Overall, the revised results strengthen the conclusion that parasitoid occurrence is influenced by host species, substrate type, habitat, trap type, and environmental conditions.

6.0. DISCUSSION

The present study synthesizes information from experimental studies on parasitoids associated with Diptera in different decomposing substrates and environments, highlighting the diversity of parasitoid species and their ecological relationships with hosts. The results demonstrate that parasitoid occurrence is strongly influenced by substrate type, host availability, environmental conditions, and sampling method. This pattern agrees with previous studies showing that decomposing organic matter, animal feces, carcasses, viscera, manure, and urban waste provide suitable resources for Diptera development and, consequently, for the establishment of their natural enemies (Marchiori, 2019d; Marchiori, 2022; Peralta-Aragón *et al.*, 2025; Rocha *et al.*, 2025; Gudin *et al.*, 2026).

The integration of data from several experimental studies allowed a broader interpretation of parasitoid–Diptera interactions than would be possible from isolated datasets. In addition to compiling biological records, the present manuscript incorporated complementary chi-square analyses to evaluate variation in parasitism rates, parasitoid abundance, substrate preference, host association, trap efficiency, habitat, and locality. This approach strengthened the interpretation of ecological patterns that were previously described mainly in qualitative or descriptive terms. Therefore, the main contribution of this synthesis is the combination of original biological information with additional statistical support for patterns involving hosts, parasitoids, substrates, and environments (Marchiori, 2022; Peralta-Aragón *et al.*, 2025; Rocha *et al.*, 2025; Gudin *et al.*, 2026).

The predominance of parasitoid species such as *B. podagrica*, *H. herbertii*, *N. vitripennis*, *P. vindemmiae*, *Spalangia*, *Trichopria*, and related taxa reflects their wide distribution and adaptability to different ecological conditions. These parasitoids are frequently associated with synanthropic Diptera and may occur in rural, urban, pasture, agroforestry, forest, poultry, and carrion-associated environments. Their presence in contrasting habitats indicates that these natural enemies are not restricted to a single ecological system, but are part of a broad network associated with decomposing substrates and immature Diptera (Marchiori, 2019a; Marchiori, 2021; Marchiori, 2022; Marchiori, 2023; Schuster and Sivakumar, 2024; Cingolani *et al.*, 2025).

Substrate type was one of the main factors influencing parasitoid diversity and parasitism rates. Carcasses, animal viscera, poultry manure, cattle dung,

buffalo dung, human feces, fish, bovine liver, and organic waste differed in their capacity to attract Diptera and sustain parasitoid activity. Substrates rich in nutrients and with high host density tended to support greater parasitoid occurrence, whereas substrates with lower host availability or less favorable decomposition conditions supported a lower parasitism rate. These differences show that parasitoid performance depends not only on the presence of the host but also on the physical and chemical conditions of the substrate, microbial activity, moisture, odor intensity, and stage of decomposition (Marchiori, 2019a; Marchiori, 2021; Marchiori, 2022; Marchiori, 2023).

Environmental factors also contributed to variation in parasitism rate among studies. Temperature, humidity, rainfall, vegetation structure, exposure time, and habitat complexity can directly affect host development, pupal availability, parasitoid searching behavior, and emergence success. Forest and agroforestry environments may provide higher microhabitat heterogeneity, while urban and livestock systems may provide continuous availability of organic substrates. In poultry farms and pasture systems, the accumulation and exposure of manure or dung can favor the development of synanthropic Diptera and create favorable conditions for parasitoid populations. These ecological differences help explain why parasitism rates and parasitoid composition varied among substrates, locations, and sampling periods (Schuster and Sivakumar, 2024; Cingolani *et al.*, 2025; Hopper and Leung, 2026; Teng *et al.*, 2026).

The occurrence of both solitary and gregarious parasitoids also helps explain differences between the number of parasitized pupae and the number of adult parasitoids obtained. Gregarious species, such as *H. herbertii* and *N. vitripennis*, may produce several individuals from a single host pupa, increasing the number of emerged parasitoids without necessarily increasing the number of parasitized pupae. In contrast, solitary parasitoids such as *P. vindemmiae* and several species of *Spalangia* usually produce one individual per host. These different reproductive strategies influence abundance estimates, parasitism rate interpretation, and the potential efficiency of parasitoids as biological control agents (Schuster and Sivakumar, 2024; Cingolani *et al.*, 2025; Hopper and Leung, 2026; Teng *et al.*, 2026).

The results also reinforce the importance of sampling methods in studies of parasitoid diversity. Metal container traps, pitfall traps, Malaise traps, yellow pan traps, baited traps, carcass exposure, and substrate-based rearing methods sample different components of the Diptera and parasitoid communities. Trap efficiency depends on insect behavior, flight activity, attraction to bait or color, proximity to host substrates, and environmental conditions. Therefore, the combined use of different sampling techniques is important for obtaining more representative data and for detecting

host–parasitoid associations that might be missed by a single method (Marchiori *et al.*, 2003c).

Overall, the synthesis confirms the ecological importance of parasitoids as natural enemies of Diptera in Brazil. Their occurrence across multiple substrates and environments demonstrates their relevance for the natural regulation of synanthropic flies of sanitary, veterinary, economic, and forensic importance. The

complementary statistical analyses added to the present manuscript reinforce that parasitoid abundance, parasitism rates, host associations, and substrate use are structured by ecological and methodological factors. Future studies should prioritize standardized sampling, accurate host and parasitoid identification, seasonal monitoring, and quantitative analyses to improve the understanding of parasitoid efficiency and their potential application in biological (Figure 42).

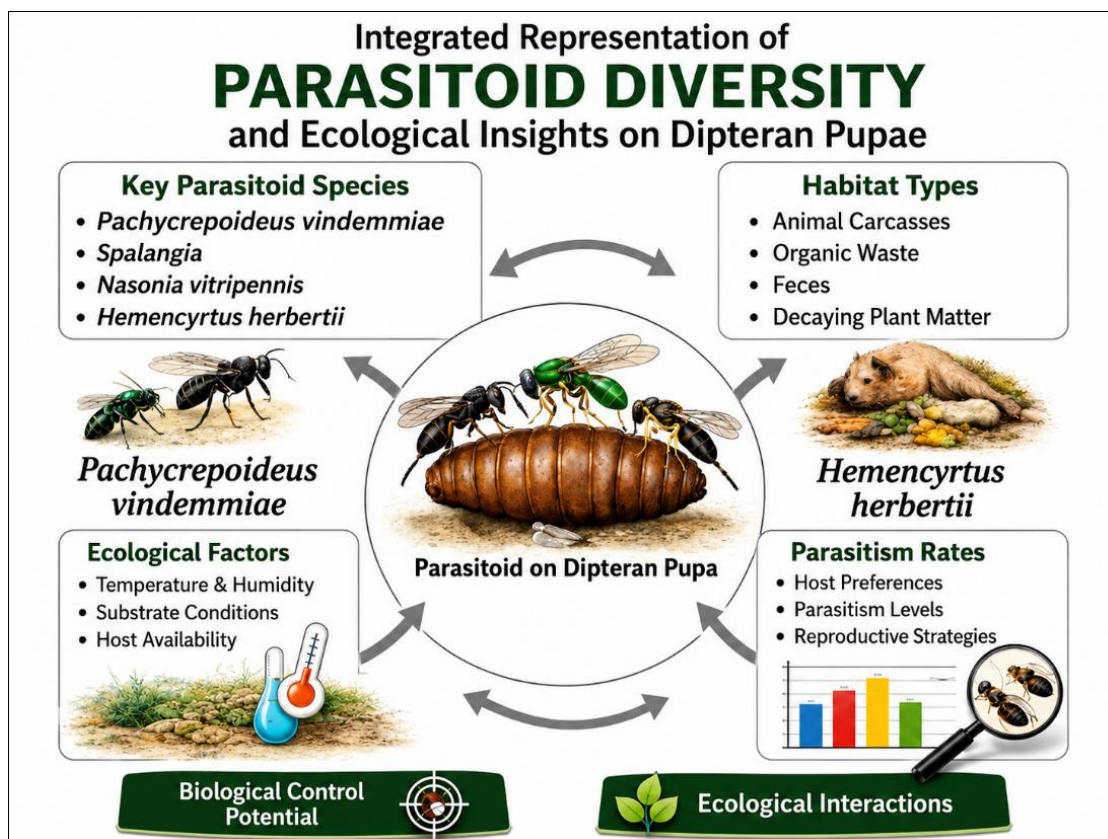


Figure 42: Integrated representation of parasitoid diversity associated with dipteran pupae. Illustration of parasitoid species, substrates, and ecological factors influencing parasitism rate. Conceptual synthesis of host–parasitoid interactions and biological control potential

7.0. CONCLUSION

The present review demonstrates that parasitoid species associated with dipteran larvae and pupae exhibit considerable diversity and ecological adaptability in different substrates and environmental conditions. The occurrence of species such as *B. podagrica*, *H. herbertii*, *N. vitripennis*, *P. vindemmiae*, and *Spalangia* highlights their importance as natural enemies of synanthropic flies. Substrate type, host availability, and environmental factors played a fundamental role in determining parasitoid distribution, parasitism rates, and host–parasitoid interactions. Environments such as carcasses, animal feces, poultry manure, and decomposing organic matter provided favorable conditions for parasitoid development, contributing to greater species richness and interaction complexity. The integration of information from tables and figures reinforced the non-random nature of parasitoid occurrence and emphasized the ecological relationships among hosts, parasitoids,

and their environments. In addition, the quantitative data available in several reviewed articles made it possible to perform complementary chi-square analyses, which were not originally presented in many of the studies. These analyses added statistical support to patterns previously described mainly in descriptive terms. Therefore, parasitoids represent an important component in the natural regulation of Diptera populations and may contribute to biological control strategies. Further studies are necessary to better understand their ecological dynamics, host specificity, seasonal variation, and potential application in integrated pest management programs.

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